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Isolation, selection, and identification of phytopathogenic fungi with bioherbicide potential for the control of field bindweed (*Convolvulus arvensis* L.)

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Abstract

Field bindweed (*Convolvulus arvensis* L.) has been described as one of the most noxious weeds in agricultural soils worldwide. Furthermore, its control by conventional herbicides can promote the emergence of resistant biotypes as well as negatively affect human health and the environment. An ecological alternative for weed management is the use of phytopathogenic fungi. In this study, 68 fungal strains were isolated from *C. arvensis* leaves exhibiting symptoms of fungal disease. Seven isolates were confirmed as pathogenic to *C. arvensis* in a detached leaf assay. Filtered cell-free cultures (FCFC) of these seven isolates were then evaluated. FCFC of TV1 and ET4 showed the best results, inhibiting seedling dry weight by up to 48% in vivo and shoot dry weight up to 35% in growth chamber assays. Phytopathogenic evaluation of both isolates showed disease severity >67%, and an ability to inhibit root dry weight by up to 80%. Inhibitory effects were not observed on chickpea, bean, sorghum, maize, or tomato plants, suggesting pathogenic specificity of TV1 and ET4 to field bindweed. Molecular identification revealed a high similarity of TV1 (100% identity) with *Macrophomina phaseolina*, and a high similarity of ET4 with *Alternaria alternata* (99.8% identity). To the best of our knowledge, this is the first report of *M. phaseolina* showing pathogenicity against field bindweed worldwide, and the first report for *A. alternata* in Mexico. The results reveal that these fungi and/or their FCFC have the potential to be used as bioherbicidal agents against *C. arvensis*.

KEYWORDS

Alternaria, *Macrophomina*, mycoherbicide, phytopathogenic fungi

1 | INTRODUCTION

Field bindweed (*Convolvulus arvensis* L.) is a highly competitive and invasive perennial weed that can persist in any type of agricultural soil (Zhang et al., 2016). This species has been described as one of the most noxious weeds in the world (Pfirter et al., 1997), and it is among the most problematic species found in agricultural soils in Mexico (Espinosa-García & Villaseñor, 2017). *Convolvulus arvensis* can reduce crop yields up to 50% by competing for water, nutrients, and space, as well as releasing allelopathic substances; it can also serve as a

reservoir for potentially pathogenic organisms and interfere with harvest procedures (Boss et al., 2007; Moura et al., 2020). Its root system can extend up to 6 m in diameter and 9 m deep (Pfirter et al., 1997), and it can produce 1×10^6 seeds per hectare, which survive up to 30 years in the soil (Timmons, 1949). These characteristics make it difficult to control by mechanical and chemical methods, and the inappropriate use of the latter can affect nontarget plants including crops of interest (Ibrahim & Tawfik, 2019). Chemical methods can also favor the appearance of resistant biotypes, as well as negatively affect the environment and human health (Harding & Raizada, 2015; Huang et al., 2019).

Due to the limitations of conventional methods, it may be necessary to develop alternative tools for weed control, such as biological products based on microorganisms (Reichert Júnior et al., 2019). Among biological agents, fungi are the most used, as they do not require a diffusion vector (Hershenhorn et al., 2016), and they are able to produce metabolites with bioherbicidal capacity (Ibrahim & Tawfik, 2019). Furthermore, they may have less of an environmental impact, which can minimize risks to human and animal health, and they have greater specificity than chemical methods (Harding & Raizada, 2015; Ibrahim & Tawfik, 2019). Therefore, the objective of this study was to isolate fungal strains from *C. arvensis* leaves, and to evaluate their bioherbicide potential on this weed.

2 | MATERIALS AND METHODS

2.1 | Isolation of phytopathogenic fungi

Fungi were isolated from *C. arvensis* plants with symptoms of foliar infection. Samples were stored in plastic bags and kept at 4°C during transport to the laboratory (Reichert Júnior et al., 2019). Selected symptomatic leaves were disinfected according to Kant et al. (2020) with some modifications. Leaves were immersed in 1% sodium hypochlorite (NaOCl) solution for 40 s, followed by washing in sterile distilled water (SDW) for 1 min. Leaves were then transferred to 30% ethanol solution for 40 s followed by three washes with SDW. Next, leaves were placed with their adaxial surface up in wet chambers made of Petri dishes containing filter paper moistened with SDW, and incubated at 25°C for 7 days to allow fungal development. After this, the fungi were subcultured three times using the hyphal fragment method (Leyronas et al., 2012) in Petri dishes with potato dextrose agar (PDA) (BD Bioxon®, Cat. No. 211900) medium in order to obtain pure cultures (Souza et al., 2017). Pure cultures were transferred to inclined test tubes containing 5 mL of PDA medium and grown at 25°C for 7 days, covered with 7 mL of sterile mineral oil and stored at room temperature.

2.2 | Bioherbicidal effect of fungi in vivo

The bioherbicide potential of the isolates was evaluated using a detached leaf assay. Leaves of *C. arvensis* were superficially disinfected using the methodology described above. Subsequently, petioles were covered with a cotton pad moistened with SDW, placed in Petri dishes, and inoculated at the adaxial surface of the leaf with a PDA disc (0.5 cm in diameter) of a 7-day old fungal isolate, using sterile PDA discs as controls (Zhang et al., 2016). The experiment was kept at 25°C for 7 days, after which foliar necrosis was evaluated (Foolad et al., 2000). Isolates showing complete colonization and necrosis of the whole leaf blade width were selected. To evaluate each isolate, three replicates with two leaves per replicate were used, and the tests were performed twice at two different times.

2.3 | Bioherbicidal effect of FCFC in vivo

2.3.1 | FCFC production

FCFC were produced according to Ibrahim and Tawfik (2019) with some modifications. Erlenmeyer flasks (500 mL) containing 200 mL of potato dextrose broth (PDB) (BD Difco™, Cat. No. 254920) were inoculated with six discs (0.5 cm in diameter) of 7-day old fungal colonies. Subsequently, the flasks were incubated at 30°C and 150 rpm for 15 days. Cultures were then centrifuged at 3350*g* for 10 min to separate cells, and the supernatant was finally filtered through a 0.45-μm nitrocellulose membrane to obtain the sterile FCFC.

2.3.2 | Bioherbicidal effect of FCFC

Seeds of *C. arvensis* and the crop plants chickpea (*Cicer arietinum*), common bean (*Phaseolus vulgaris*), sorghum (*Sorghum bicolor*), and corn (*Zea mays*) were disinfected according to Meena et al. (2016), with some modifications. The seeds were immersed in a 1% NaOCl solution for 5 min, followed by an SDW wash for 1 min. Subsequently, 30% ethanol was added for 3 min, and the seeds were finally washed three times with SDW. Seeds were transferred to a Petri dish with sterile absorbent paper and inoculated with 5 mL of FCFC of each selected isolate. The plates were incubated in the dark at 25°C for 7 days. Subsequently, germination percentage, seedling

length and dry weight data were collected. The test was repeated twice with three replicates per treatment and 20 seeds per replicate. Isolates that had the greatest bioherbicidal effect on weeds without affecting the crops were selected.

2.4 | Bioherbicidal effect of fungi in the growth chamber

2.4.1 | Plant growth conditions

Convolvulus arvensis, chickpea, common bean, sorghum, corn, as well as tomato (*Solanum lycopersicum*) seeds, were disinfected and sown 3 cm deep in polystyrene germination trays (3.175 cm × 3.175 cm × 6.35 cm) containing a mixture of sand/vermiculite substrate (1:2 v/v; sterilized by autoclaving at 121°C and 15 PSI for 1 h, repeated for three consecutive days and stored at room temperature for at least 1 day before use). Trays were placed in a growth chamber (Luceren®, Model RTOP-1000D) at 60% relative humidity with a 16-h light and 8-h dark photoperiod at 28 and 16°C, respectively. Plants were irrigated with SDW every third day and fertilized once per week with Hoagland's solution (Hoagland & Arnon, 1950).

2.4.2 | Bioherbicide effect of fungi isolates

C. arvensis plants in the developmental stage with three true leaves and 20-day old plants of chickpea, common bean, sorghum, corn and tomato received wounds on all true leaves by puncturing them with a sterile needle to favor infection. Fungal inoculum was produced by growing in PDB medium each isolate, producing different fungal structures: *M. phaseolina* produced microsclerotia and mycelium and *A. alternata* conidia and mycelium. Colony forming units (CFU) per mL was quantitated per each isolate in order to count the infective units present in each inoculum. Plants were individually sprayed with the isolates selected in vivo using 1 mL of fungal structures suspended in 0.1% Tween 20 (Azumex®) at a concentration of 1×10^6 CFU mL^{-1}, with SDW used as a control. Once inoculated, plants were covered with clear polyethylene bags for 48 h to retain moisture. The growth chamber trays were kept for 21 days for *C. arvensis* and 15 days for crop plants under the aforementioned conditions.

The severity of disease caused by pathogens was analyzed by calculating the Disease Severity Index (DSI) using Equation (1) (Townsend & Heuberger, 1943), based on the scale proposed by Razaghi and Zafari (2017). This scale consists of five levels, where 0 = no symptoms, 1 = less than 10% of the plant surface is covered by lesions, 2 = 10%–25% coverage, 3 = 25%–50% coverage, 4 = 50%–75% coverage, and 5 = 75%–100% coverage, resulting in the total death of the plant.

$$\mathrm{DSI}\% = \frac{\sum(n \times v)}{z \times N} \times 100, \quad (1)$$

where n = number of plants in each scale; v = scale value; z = highest scale value; and N = total number of plants.

Bioassays were performed twice independently using a completely randomized design with 10 plants per replicate and six replicates per treatment. In addition, disease incidence, plant length, and biomass were measured. To comply with Koch's postulates, fungi were isolated from symptomatic *C. arvensis* plants from the bioassay, and their identity was confirmed based on morphological traits and molecular identification (Abdessemed, Kerroum, Bahet, Talbi, & Zermane, 2019).

2.5 | Effect of FCFC in the growth chamber

Seeds of *C. arvensis*, chickpea, common bean, sorghum, corn, and tomato crops were disinfected and planted in polystyrene germination trays, following the plant growth conditions described above. Next, 6 mL of FCFC from each isolate selected in the in vivo assays were added to each cell, with PDB medium used as a control. The trays were placed in a growth chamber under the aforementioned conditions, and the plants were watered with distilled water every third day and fertilized once per week with Hoagland's solution (Hoagland & Arnon, 1950). Fifteen days after inoculation (DAI), the germination percentage, seedling length, and biomass data were collected. The bioassays were performed twice, using a completely randomized design with one seed per cell, six cells per replicate, and three replicates per treatment (Verdugo-Navarrete et al., 2021).

2.6 | Molecular identification

2.6.1 | DNA extraction

Fungal isolates were grown for 7 days in PDA medium, after which 10 mg of fungal mycelium or 50 mg of *C. arvensis* leaf tissue were collected for DNA extraction. DNA was extracted using DNAzol (Research Center Inc., Cat. No. DN 127) following the manufacturer's

specifications. The quality of the DNA was monitored in a NanoDrop 2000_C spectrophotometer (Thermo Scientific), and by visualization using agarose gel electrophoresis.

2.6.2 | DNA amplification and sequencing

The selected isolates and the initially unknown *C. arvensis* weed were identified by sequencing the ITS rDNA region, using the oligonucleotides ITS1 (5′-TCCGTAGGTGAACCTGCGG-3′) and ITS4 (5′-TCCTCCGCTTATTGATATGC-3′) (White et al., 1990). Similarly, glyceraldehyde-3-phosphate dehydrogenase (GAPDH) and translation elongation factor 1-α (TEF1-α) were identified using the primer pairs gpd1 (5′-CAACGGCTTCGGTCGCATTG-3′) and gpd2 (5′-GCCAAGCAGTTGGTTGTGC-3′) (Berbee et al., 1999), and EF1-728F (5′-CATCGAGAAGTTCGAGAAGG-3′) and EF1-986R (5′-TACTTGAAGGAACCCTTACC-3′) (Carbone & Kohn, 1999), respectively. The PCR mix contained 1 μL (50–100 ng) of DNA template, 1X of reaction buffer, 1.5 mM of $MgCl_2$, 0.4 mM of each primer, 500 μM of deoxynucleotide triphosphate (dNTP) (Sigma-Aldrich, Cat. No. U1240, Madison, WI) and 1.25 U of GoTaq DNA polymerase (Promega, Cat. No. M8295, Madison, WI) in a total volume of 25 μL. The PCR was performed using a thermal cycler (Labnet, Cat. No. TC9600-G). The conditions to amplify the ITS rDNA region consisted of an initial denaturation step with one cycle of 94°C for 3 min, followed by 35 cycles of 94°C for 30 s, 60°C for 30 s, 72°C for 1 min, and a final extension step of 72°C for 5 min. The conditions for GAPDH amplification consisted of an initial denaturation step of 95°C for 2 min, followed by 35 cycles of 95°C for 1 min, 52°C for 1 min, 72°C for 45 s, and a final extension of 72°C for 10 min. For TEF1-α, an initial denaturation temperature of 94°C for 5 min was used, followed by 35 cycles of 94°C for 30 s, 55°C for 30 s, 72°C for 1 min, and a final extension of 72°C for 5 min. The amplified products were purified using the commercial QIAquick® PCR Purification Kit (QIAGEN, Cat. No. 28106, Hilden, Germany) according to the manufacturer's specifications. The PCR products were sequenced bidirectionally at the National Genomics and Biodiversity Laboratory (LANGEBIO, Irapuato, Mexico). Sequences were edited in the Bio Edit program and compared to sequences in the NCBI (National Center for Biotechnology Information) website using the BLAST-N software.

2.7 | Statistical analysis

The data of germination percentage, incidence and DSI were arcsine–transformed. All results were subjected to analysis of variance (ANOVA) and Tukey's test ($p < .05$). Statistical differences between treatments and controls in the different cultures were calculated using Student's *t*-test. Data were analyzed using the SPSS software (IBM SPSS Statistics for Windows, version 25.0. IBM, Armonk, New York, NY).

3 | RESULTS

3.1 | Isolation and purification of phytopathogenic fungi

A total of 68 fungal isolates were obtained from 30 *C. arvensis* plant samples with symptoms of fungal diseases. The plants were collected from chickpea, bean and corn crop fields in the northern region of the State of Sinaloa, Mexico, from December 2019 to March 2020.

3.2 | Evaluation of phytopathogenic fungi in vivo

Leaves treated with the isolates GU18, TV17, TV22, ET4, EN9, N8, and TV1 showed serious symptoms of disease in two independent experiments. The symptomatology consisted of the presence of brown necrotic spots and chlorosis, as well as the complete mycelial colonization of the leaf blade width 7 DAI (Figure 1).

3.3 | Effect of FCFC in vivo

Once the detached leaf assay was performed, isolates GU18, TV1, TV17, TV22, ET4, EN9, and N8 were selected to measure the effect of FCFC on the development of *C. arvensis* seedlings.

The filtrates showed inhibition of *C. arvensis* seedling development, although no effect on germination was observed with any of the isolates (Figure 2 and Table 1). The FCFC of TV1, TV22, ET4, and EN9 inhibited seedling length by 87%, 81%, 80%, and 82%, respectively. These filtrates also showed a decrease in *C. arvensis* dry weight by 48%, 42%, 47%, and 40%, respectively (Table 1).

The in vivo assay of FCFC in chickpea, sorghum, common bean, and corn showed that isolate GU18 affected length and dry weight in chickpea, bean and corn seedlings, but not in sorghum. While TV1 filtrate did not inhibit the development of any of the crops, isolate TV17 affected chickpea, bean and corn seedling length, in addition to corn biomass. TV22 affected the

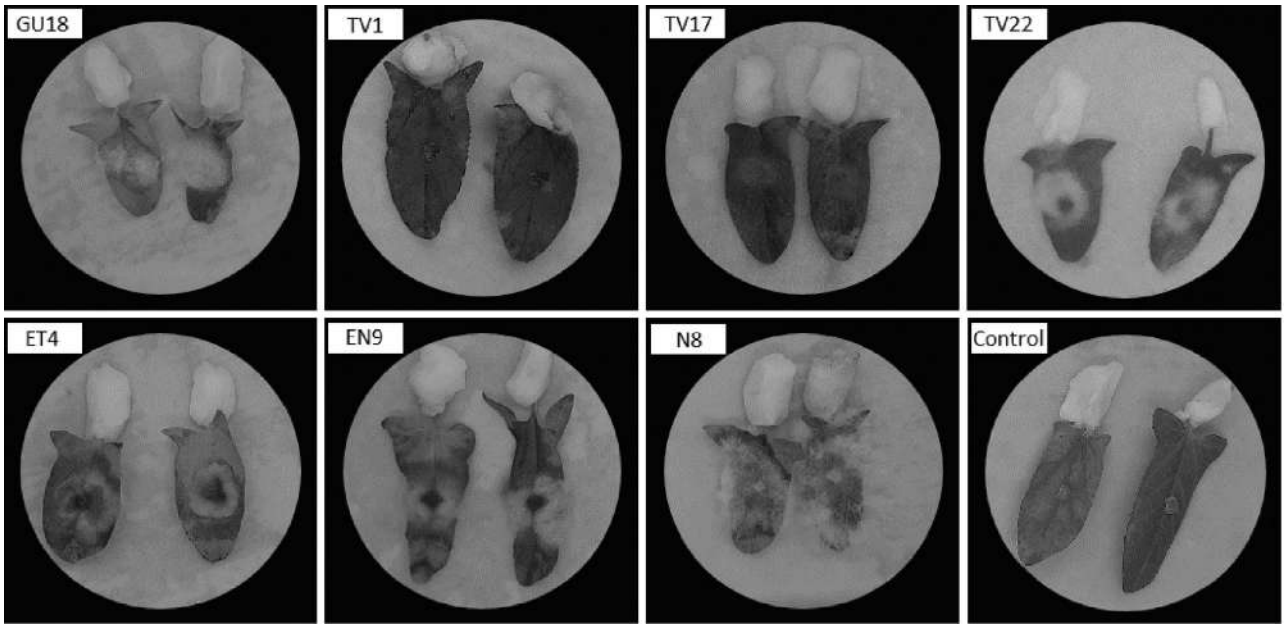

FIGURE 1 Effect of fungal isolates on *Convolvulus arvensis* leaves in vivo 7 days after inoculation. Control refers to the addition of a potato dextrose agar disc without fungus.

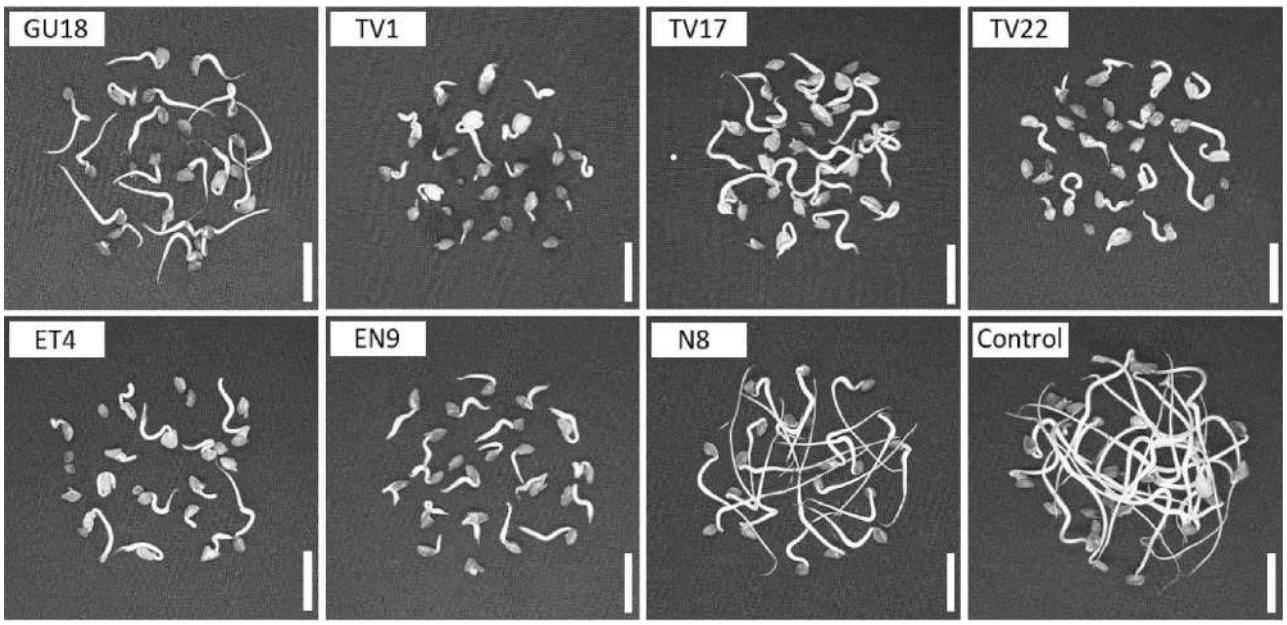

FIGURE 2 Effect of filtered cell-free cultures from fungal isolates on *Convolvulus arvensis* seed germination and seedling development in vivo at 7 days after inoculation. Control refers to a potato dextrose broth treatment without fungal filtered cell-free cultures. Scale bar = 2 cm.

length and dry weight of sorghum, and length development in chickpea and bean, in addition to decreasing corn biomass. ET4 filtrate caused a decrease in the length of bean and corn plants, without affecting the development of chickpea and sorghum. EN9 and N8 filtrates caused a decrease in the length of chickpea and bean plants, without affecting the development of sorghum, although they did affect corn biomass (Table S1). The effect of FCFC in vivo on *C. arvensis* and crop plants was consistently observed in the two independent experiments.

Due to their capacity to inhibit *C. arvensis* in vivo, strains TV1 and ET4 were selected to conduct tests in a growth chamber.

Isolate	Length (cm)	Dry weight (mg)	Germination (%)
GU18	3.30 ± 0.09^{d}	5.21 ± 0.24^{c}	96.67 ± 5.77^{a}
TV1	1.16 ± 0.03^{a}	3.62 ± 0.04^{a}	91.67 ± 2.88^{a}
TV17	2.66 ± 0.11^{c}	5.67 ± 0.09^{c}	95.00 ± 8.66^{a}
TV22	1.69 ± 0.04^{b}	4.06 ± 0.06^{ab}	95.00 ± 8.66^{a}
ET4	1.74 ± 0.07^{b}	3.74 ± 0.21^{ab}	80.00 ± 10.00^{a}
EN9	1.55 ± 0.11^{b}	4.18 ± 0.17^{b}	95.00 ± 8.66^{a}
N8	5.12 ± 0.15^{e}	5.40 ± 0.26^{c}	100.00 ± 0.00^{a}
Control	8.85 ± 0.26^{f}	7.02 ± 0.14^{d}	90.00 ± 10.00^{a}

Note: Data followed by different superscript letters in a column differ significantly ($p \leq .05$). Control refers to the treatment with potato dextrose broth.

TABLE 1 Effect of filtered cell-free cultures on the development and germination of *Convolvulus arvensis* seedlings in vivo.

3.4 | Plant pathogenicity test

C. arvensis plants inoculated with isolates TV1 and ET4 showed a disease incidence of 100%. Necrotic spots and chlorosis in cotyledons were visible 3 to 4 DAI, and symptomatic areas expanded into true leaves and stems 5 to 6 DAI. Disease development resulted in partial colonization of treated and emerging shoots (Figure 3 and Figure S1), resulting in DSI values of 88% and 67% for TV1 and ET4, respectively (Table 2). The inoculated pathogens were re-isolated to accomplish Koch's postulates and their identity was confirmed by morphological and molecular methods, thus finding similarity with the original isolates.

Isolate TV1 significantly inhibited *C. arvensis* root length during its development by 46% in comparison to the control. Isolates TV1 and ET4 inhibited shoot length by 57% and 49%, respectively, and decreased root dry weight by 80% and 58%. The highest inhibition of shoot dry weight was observed with isolate TV1 (64%) followed by treatment with ET4 (51%), as compared to the control (Table 2).

Isolates TV1 and ET4 did not exhibit any disease symptoms or negative effects on the development of chickpea, bean, sorghum, corn, or tomato plants (Table S2 and Figure S2). The effect of these isolates was consistent in the two independent experiments.

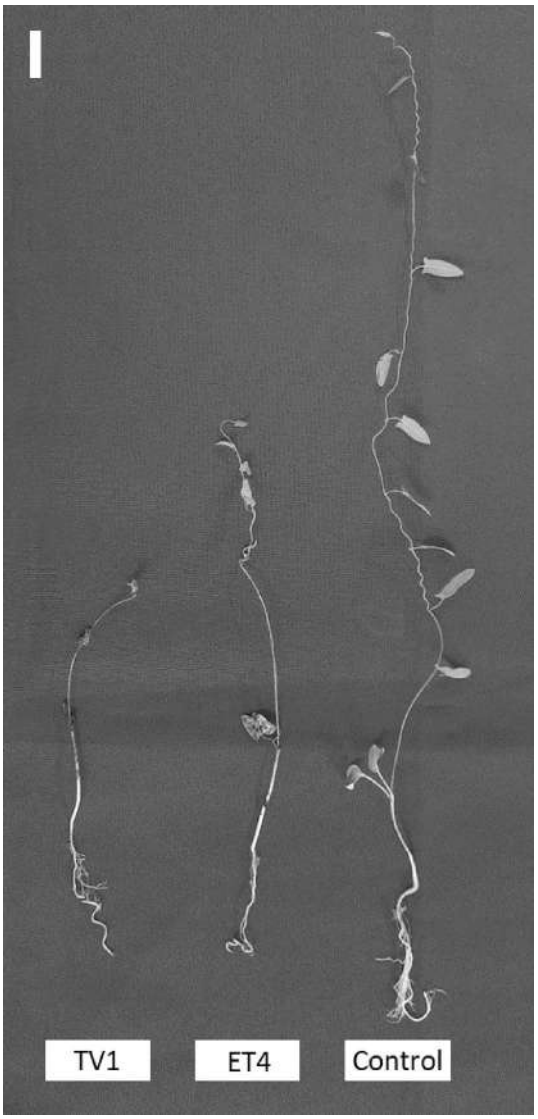

FIGURE 3 Effect of fungal isolates TV1 and ET4 on *Convolvulus arvensis* plants 21 days after inoculation. Control refers to treatment with sterile distilled water. Scale bar = 2 cm.

3.5 | Evaluation of FCFC in the growth chamber

The filtrates of strains TV1 and ET4 induced a decrease in the development of *C. arvensis* seedlings (Figure 4 and Figure S3), with root length inhibition ranging from 21% to 26% and shoot inhibition ranging from 63% to 69% (Table 3). No difference in dry weight was observed between treatments and control in the roots; however, shoot

TABLE 2 Effect of phytopathogenic fungi on *Convolvulus arvensis* development, incidence, and disease severity in the growth chamber.

	Length (cm)		Dry weight (mg)			
Isolate	**Root**	**Shoot**	**Root**	**Shoot**	**Incidence (%)**	**DSI (%)**
TV1	4.89 ± 1.00[a]	16.93 ± 2.02[a]	3.20 ± 0.93[a]	16.83 ± 2.81[a]	100.00 ± 0.00[b]	88.00 ± 5.66[c]
ET4	7.20 ± 0.82[b]	19.93 ± 1.54[a]	6.70 ± 0.90[a]	23.15 ± 2.19[b]	100.00 ± 0.00[b]	67.00 ± 11.49[b]
Control	9.05 ± 1.52[b]	39.02 ± 1.52[b]	16.07 ± 4.49[b]	47.38 ± 4.46[c]	0.00 ± 0.00[a]	0.00 ± 0.00[a]

Note: Data followed by different superscript letters in a column differ significantly ($p \leq .05$). Control refers to the treatment with sterile distilled water.
Abbreviation: DSI, Disease Severity Index.

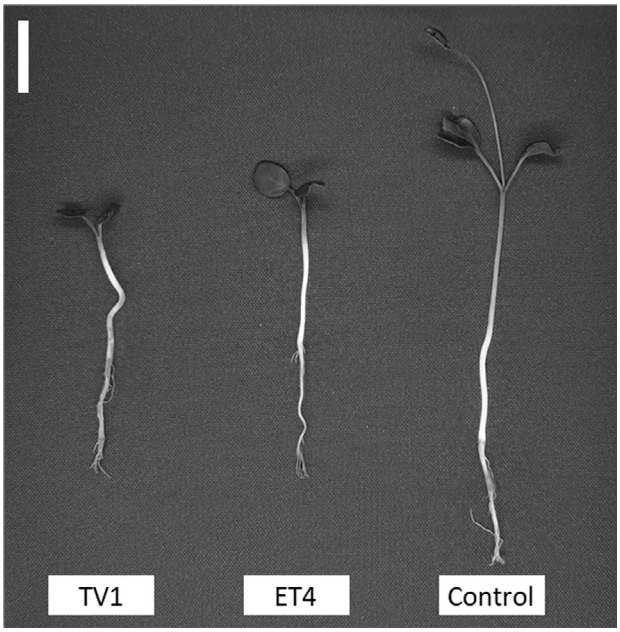

FIGURE 4 Effect of filtered cell-free cultures from TV1 and ET4 isolates on the germination and development of *Convolvulus arvensis* 15 days after inoculation. Control refers to a potato dextrose broth treatment without fungal filtered cell-free cultures. Scale bar = 2 cm.

inhibition ranged from 30% to 35% (Table 3). As in the in vivo results, no germination inhibition was observed (Table 3).

The filtrates did not affect the development of chickpea, bean, sorghum, corn, or tomato plants (Table S3 and Figure S4). The effect of FCFC in the growth chamber on *C. arvensis* and crop plants was consistent in the two independent experiments.

3.6 | Molecular identification

The PCR of the ITS region of isolates GU18, TV1, TV17, TV22, ET4, EN9, N8, and *C. arvensis* yielded products of ~650 bp. The sequences obtained were compared with those reported in the database of the National Center for Biotechnology Information (NCBI). According to the BLAST-N results (100% identity, 100% coverage, and value $E = 0$), the isolates belong to the genera *Macrophomina* (TV1), *Alternaria* (TV17, TV22, ET4, and EN9), and *Fusarium* (GU18 and N8). Initially unknown, the weed was identified as *C. arvensis* (100% identity, 100% coverage, and value $E = 0$). The obtained ITS sequence of *C. arvensis* was registered in the GenBank under the accession number OP286632.

Isolate TV1 was identified at the species level by amplification and sequencing of the TEF1-α gene (350 bp), while isolate ET4 was identified by amplification and sequencing of the GAPDH gene (600 bp). TV1 sequences showed and an E value =0, 100% identity, and 99% coverage with *M. phaseolina*, while ET4 sequences showed an E value = 0, 99.8% identity, and 100% coverage with *Alternaria alternata*. Both sequences, EF1-α of isolate TV1 and GAPDH of isolate ET4, were registered in the GenBank under the accession numbers OP296502 and OP296501, respectively.

4 | DISCUSSION

The objective of this work was to obtain fungal isolates capable of inhibiting the development of the weed *C. arvensis*. Isolates TV1 (*M. phaseolina*) and ET4 (*A. alternata*) and their FCFC were selected for their bioherbicidal effect.

Although biological control of *C. arvensis* by phytopathogenic fungi has already been reported (El-Sayed & Hurle, 2001; Ibrahim & Tawfik, 2019; Pfirter et al., 1997), there are no reports of *M. phaseolina* phytopathogenicity on *C. arvensis*. In this study, the phytotoxic effect of *M. phaseolina* on the host plant could be related to the production of cell wall hydrolytic enzymes (Marquez et al., 2021), which could support the entry of the biological agent or phytotoxins produced by it (Ghorbani et al., 2005; Harding & Raizada, 2015; Reichert Júnior et al., 2019). Such phytotoxins include phaseolinon, botryodiplodin, and patulin, which have been reported to play an important role in the initial stage of infection in *M. phaseolina*, causing wilting of seedlings and necrotic

TABLE 3 Effect of filtered cell-free cultures on development and germination of *Convolvulus arvensis* seedlings in the growth chamber.

	Length (cm)		Dry weight (mg)		
Isolate	**Root**	**Shoot**	**Root**	**Shoot**	**Germination (%)**
TV1	4.02 ± 0.35[a]	3.82 ± 0.39[a]	3.48 ± 0.39[a]	8.36 ± 0.39[a]	83.33 ± 28.87[a]
ET4	4.27 ± 0.30[a]	3.24 ± 0.16[a]	3.31 ± 0.45[a]	9.05 ± 0.48[a]	72.22 ± 19.24[a]
Control	5.40 ± 0.05[b]	10.37 ± 0.04[b]	3.32 ± 0.40[a]	12.90 ± 0.40[b]	94.44 ± 9.62[a]

Note: Data followed by different superscript letters in a column differ significantly ($p \leq .05$). Control refers to treatment with potato dextrose broth.

lesions on leaves and roots (Abbas et al., 2020; Bhattacharya et al., 1987; Marquez et al., 2021; Salvatore et al., 2020).

C. arvensis can be naturally infected by *A. alternata* (Abdessemed et al., 2021), as is the case for isolate ET4. *Alternaria* species have been evaluated for their potential as mycoherbicides against weeds such as *Parthenium hysterophorus* (Abdessemed et al., 2020; Saxena & Kumar, 2007, 2010), *Xanthium strumarium* (Abdessemed et al., 2020; Abdessemed, Kerroum, Bahet, & Zermane, 2019), *Parthenium* sp. (Meena et al., 2016), and *Amaranthus retroflexus* (Ghorbani et al., 2000; Lawrie et al., 2000, 2001; Lawrie et al., 2002; Qiang et al., 2006).

The bioherbicidal effect of *A. alternata* could be due to its ability to produce tenuazonic acid. This phytotoxin is produced by members of the *Alternaria* genus (Chen & Qiang, 2017; Ismaiel & Papenbrock, 2015) and has the potential to inhibit the activity of *p*-hydroxyphenylpyruvate dioxygenase (Meazza et al., 2002), affecting shoot and root elongation of seedlings (Chen & Qiang, 2017; Marfori et al., 2003; Tylkowska et al., 2003; Zonno & Vurro, 1999). Furthermore, tenuazonic acid can interrupt the electron transport of photosystem II (PSII) (Chen & Qiang, 2017), which favors the formation of reactive oxygen species (ROS) (Apel & Hirt, 2004; Laloi et al., 2004), causing cell destruction and leaf necrosis (Chen & Qiang, 2017; Wan et al., 2001).

In the present study, the application of isolates TV1 and ET4, as well as their filtrates, decreased the development of *C. arvensis* without affecting chickpea, bean, sorghum, corn, or tomato (a crop close to *Convolvulaceae* family), which demonstrates their potential as bioherbicidal agents. The specificity of mycoherbicides has already been observed in *A. alternata* (Abdessemed et al., 2020; Babu et al., 2003; Kotan et al., 2013; Siddiqui et al., 2010), which may be attributable to specific host toxins derived from some fungi (Abdessemed et al., 2020; Meena & Samal, 2019; Tsuge et al., 2013), in combination with phytohormones produced by these microorganisms that could have inhibitory or growth-promoting effects depending on the metabolite (i.e., toxin/phytohormone) concentration, of the species plant, the physicochemical characteristics of the soil, the presence of other microorganisms, and so forth (Nehl et al., 1997; Sindhu et al., 2018; Verdugo-Navarrete et al., 2021).

The growth inhibition of *C. arvensis* caused by the TV1 and ET4 filtrates was lower in the growth chamber assays than in the in vivo tests. Indeed, the metabolites produced by these isolates may have a short half-life (Li et al., 2003). It is also possible that periodic watering diluted the filtrates, lowering the inhibitory effect of the filtrates. This phenomenon was previously reported by Zhou et al. (2019), who found that the effect of the secondary metabolites of *A. alternata* in the plant is greater when their concentration increases.

The level of disease and weed inhibition by phytopathogenic fungi could be increased by combining the filtrates and/or pathogens (Abdessemed et al., 2021). While a bioherbicide is not expected to show the same effect on plants as synthetic herbicides, bioherbicides can provide a competitive advantage for crop seedlings through weed infection and seedling growth retardation (Souza et al., 2017). In order to improve the bioherbicidal effect, formulations and integrated management approaches must be developed. Importantly, bioherbicide development may contribute to improving sustainable agriculture practices (Boyetchko et al., 2002; Meena et al., 2016; Pfirter et al., 1997).

Although *A. alternata* has been shown to act as a pathogen of *C. arvensis* in other countries, this is the first report to describe this interaction in Mexico. This is also the first report of *M. phaseolina* showing pathogenicity against *C. arvensis* anywhere in the world, to the best of our knowledge. The fungal isolates and FCFC of TV1 (*M. phaseolina*) and ET4 (*A. alternata*) were selected out of 68 strains for their in vivo bioherbicidal effect. In growth chamber assays, these isolates inhibited *C. arvensis* development, without affecting the development of crops of agricultural interest. Since the metabolites present in FCFC should be important for their phytotoxicity effects, it is possible to visualize a management strategy involving the combination of filtrates and/or fungal agents as well as their formulations in order to maximize the bioherbicidal effect. Further

assessments should be carried out under greenhouse and open field conditions to confirm *C. arvensis* control. It is also advisable to expand testing on various crops of interest to confirm the absence of negative effects on plant development and to validate the specificity of the selected isolates. Finally, future studies should aim to conduct metabolomic studies to elucidate the possible compounds involved in the bioherbicidal mechanisms used by TV1 and ET4 on *C. arvensis*.

AUTHOR CONTRIBUTIONS

Sotelo-Cerón, N. D., Maldonado-Mendoza I. E., Leyva-Madrigal, K. Y. and Martínez-Álvarez, J. C. contributed equally to all stages of the study and preparation of this article.

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CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest.

DATA AVAILABILITY STATEMENT

All data generated or analyzed during this study are included in this published article and its supplementary information files.

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REFERENCES

Abbas, H. K., Bellaloui, N., Butler, A. M., Nelson, J. L., Abou-Karam, M., & Shier, W. T. (2020). Phytotoxic responses of soybean (*Glycine max* L.) to botryodiplodin, a toxin produced by the charcoal rot disease fungus, *Macrophomina phaseolina*. *Toxins*, *12*(1), 25. https://doi.org/10.3390/toxins12010025

Abdessemed, N., Bahet, Y. A., & Zermane, N. (2020). Mycoherbicide potential of *Alternaria alternata* (Fries.) Kiessler and its formulations on the host weed *Xanthium strumarium* L. *Biocontrol Science and Technology*, *30*(12), 1300–1315. https://doi.org/10.1080/09583157.2020.1814692

Abdessemed, N., Kerroum, A., Bahet, Y., Talbi, N., & Zermane, N. (2019). First report of *Alternaria* leaf spot caused by *Alternaria alternata* (Fries.) Kiessler on *Sonchus oleraceus* L. and *Convolvulus arvensis* L. in Algeria. *Journal of Phytopathology*, *167*(6), 321–325. https://doi.org/10.1111/jph.12800

Abdessemed, N., Kerroum, A., Bahet, Y. A., & Zermane, N. (2019). First report of *Alternaria alternata* leaf spot on *Xanthium strumarium* L. in Algeria. *Journal of Plant Pathology*, *101*(3), 781. https://doi.org/10.1007/s42161-019-00245-2

Abdessemed, N., Staropoli, A., Zermane, N., & Vinale, F. (2021). Metabolic profile and mycoherbicidal activity of three *Alternaria alternata* isolates for the control of *Convolvulus arvensis*, *Sonchus oleraceus*, and *Xanthium strumarium*. *Pathogens*, *10*(11), 1–11. https://doi.org/10.3390/pathogens10111448

Apel, K., & Hirt, H. (2004). Reactive oxygen species: Metabolism, oxidative stress, and signal transduction. *Annual Review of Plant Biology*, *55*(1), 373–399. https://doi.org/10.1146/annurev.arplant.55.031903.141701

Babu, R. M., Sajeena, A., & Seetharaman, K. (2003). Bioassay of the potentiality of *Alternaria alternata* (Fr.) Keissler as a bioherbicide to control waterhyacinth and other aquatic weeds. *Crop Protection*, *22*(8), 1005–1013. https://doi.org/10.1016/S0261-2194(03)00115-7

Berbee, M. L., Pirseyedi, M., & Hubbard, S. (1999). *Cochliobolus* phylogenetics and the origin of known, highly virulent pathogens, inferred from ITS and glyceraldehyde-3-phosphate dehydrogenase gene sequences. *Mycologia*, *91*(6), 964–977. https://doi.org/10.1080/00275514.1999.12061106

Bhattacharya, G., Dhar, T. K., Bhattacharyya, F. K., & Siddiqui, K. A. (1987). Mutagenic action of phaseolinone, a mycotoxin isolated from *Macrophomina phaseolina*. *Australian Journal of Biological Sciences*, *40*(4), 349–353. https://doi.org/10.1071/bi9870349

Boss, D., Schläpfer, E., Fuchs, J., Défago, G., & Maurhofer, M. (2007). Improvement and application of the biocontrol fungus *Stagonospora convolvuli* LA39 formulation for efficient control of *Calystegia sepium* and *Convolvulus arvensis*. *Journal of Plant Diseases and Protection*, *114*(5), 232–238. https://doi.org/10.1007/BF03356223

Boyetchko, S. M., Rosskopf, E. N., Caesar, A. J., & Charudattan, R. (2002). Biological weed control with pathogens: Search for candidates to applications. In G. G. Khachatourians & D. K. Arora (Eds.), *Applied mycology and biotechnology* (Vol. 2, pp. 239–274). Elsevier. https://doi.org/10.1016/S1874-5334(02)80013-2

Carbone, I., & Kohn, L. M. (1999). A method for designing primer sets for speciation studies in filamentous ascomycetes. *Mycologia*, *91*(3), 553–556. https://doi.org/10.1080/00275514.1999.12061051

Chen, S., & Qiang, S. (2017). Recent advances in tenuazonic acid as a potential herbicide. *Pesticide Biochemistry and Physiology*, *143*, 252–257. https://doi.org/10.1016/j.pestbp.2017.01.003

El-Sayed, W., & Hurle, K. (2001). Efficacy of *Phomopsis convolvulus* as a mycoherbicide for *Convolvulus arvensis*. *Mededelingen (Rijksuniversiteit te Gent. Fakulteit van de Landbouwkundige en Toegepaste Biologische Wetenschappen)*, *66*, 775–789.

Espinosa-García, F. J., & Villaseñor, J. L. (2017). Biodiversity, distribution, ecology and management of non-native weeds in Mexico: A review. *Revista Mexicana de Biodiversidad*, *88*, 76–96. https://doi.org/10.1016/j.rmb.2017.10.010

Foolad, M. R., Ntahimpera, N., Christ, B. J., & Lin, G. Y. (2000). Comparison of field, greenhouse, and detached-leaflet evaluations of tomato germ plasm for early blight resistance. *Plant*

Disease, *84*(9), 967–972. https://doi.org/10.1094/PDIS.2000.84.9.967

Ghorbani, R., Leifert, C., & Seel, W. (2005). Biological control of weeds with antagonistic plant pathogens. *Advances in Agronomy*, *86*, 191–225. https://doi.org/10.1016/S0065-2113(05)86004-3

Ghorbani, R., Seel, W., Litterick, A., & Leifert, C. (2000). Evaluation of *Alternaria alternata* for biological control of *Amaranthus retroflexus*. *Weed Science*, *48*(4), 474–480. https://doi.org/10.1614/0043-1745(2000)048[0474:EOAAFB]2.0.CO;2

Harding, D. P., & Raizada, M. N. (2015). Controlling weeds with fungi, bacteria and viruses: A review. *Frontiers in Plant Science*, *6*(659), 1–14. https://doi.org/10.3389/fpls.2015.00659

Hershenhorn, J., Casella, F., & Vurro, M. (2016). Weed biocontrol with fungi: Past, present and future. *Biocontrol Science and Technology*, *26*(10), 1313–1328. https://doi.org/10.1080/09583157.2016.1209161

Hoagland, D. R., & Arnon, D. I. (1950). *The water-culture method for growing plants without soil* (Vol. 347, 2nd ed.). College of Agriculture, University of California.

Huang, Z., Liu, Y., Zhang, C., Jiang, C., Huang, H., & Wei, S. (2019). Molecular basis of natural tolerance to glyphosate in *Convolvulus arvensis*. *Scientific Reports*, *9*, 8133. https://doi.org/10.1038/s41598-019-44583-8

Ibrahim, N., & Tawfik, M. (2019). Fungi as potential biocontrol agents against *Convolvulus arvensis* and *Portulaca oleracea* infesting the agroecosystems of Egypt. *Egyptian Journal of Microbiology*, *54*(1), 117–135. https://doi.org/10.21608/ejm.2019.17443.1117

Ismaiel, A. A., & Papenbrock, J. (2015). Mycotoxins: Producing fungi and mechanisms of phytotoxicity. *Agriculture*, *5*(3), 492–537. https://doi.org/10.3390/agricultura5030492

Kant, R., Joshi, P., Bhandari, M. S., Pandey, A., & Pandey, S. (2020). Identification and pathogenicity of *Alternaria alternata* causing leaf spot and blight disease of *Ailanthus excelsa* in India. *Forest Pathology*, *50*(2), e12584. https://doi.org/10.1111/efp.12584

Kotan, R., Okutucu, A., Ala Görmez, A., Karagoz, K., Dadasoglu, F., Karaman, İ., Hasanekoglu, İ., & Kordali, Ş. (2013). Parasitic bacteria and fungi on common mistletoe (*Viscum album* L.) and their potential application in biocontrol. *Journal of Phytopathology*, *161*(3), 165–171. https://doi.org/10.1111/jph.12048

Laloi, C., Apel, K., & Danon, A. (2004). Reactive oxygen signalling: The latest news. *Current Opinion in Plant Biology*, *7*(3), 323–328. https://doi.org/10.1016/j.pbi.2004.03.005

Lawrie, J., Down, V. M., & Greaves, M. P. (2000). Factors influencing the efficacy of the potential microbial herbicide *Alternaria alternata* (Fr.) Keissler on *Amaranthus retroflexus* (L.). *Biocontrol Science and Technology*, *10*(1), 81–87. https://doi.org/10.1080/09583150029413

Lawrie, J., Down, V. M., & Greaves, M. P. (2001). Effects of storage on viability and efficacy of granular formulations of the microbial herbicides *Alternaria alternata* and *Trematophoma lignicola*. *Biocontrol Science and Technology*, *11*(2), 283–295. https://doi.org/10.1080/09583150120035701

Lawrie, J., Greaves, M. P., Down, V. M., Morales-Aza, B., & Lewis, J. M. (2002). Outdoor studies of the efficacy of *Alternaria alternata* in controlling *Amaranthus retroflexus*. *Biocontrol Science and Technology*, *12*(1), 83–94. https://doi.org/10.1080/09583150120110671

Leyronas, C., Duffaud, M., & Nicot, P. C. (2012). Compared efficiency of the isolation methods for *Botrytis cinerea*. *Mycology*, *3*(4), 221–225. https://doi.org/10.1080/21501203.2012.727484

Li, Y., Sun, Z., Zhuang, X., Xu, L., Chen, S., & Li, M. (2003). Research progress on microbial herbicides. *Crop Protection*, *22*(2), 247–252. https://doi.org/10.1016/S0261-2194(02)00189-8

Marfori, E. C., Kajiyama, S. I., Fukusaki, E., & Kobayashi, A. (2003). Phytotoxicity of the tetramic acid metabolite trichosetin. *Phytochemistry*, *62*(5), 715–721. https://doi.org/10.1016/S0031-9422(02)00629-5

Marquez, N., Giachero, M. L., Declerck, S., & Ducasse, D. A. (2021). *Macrophomina phaseolina*: General characteristics of pathogenicity and methods of control. *Frontiers in Plant Science*, *12*, 634397. https://doi.org/10.3389/fpls.2021.634397

Meazza, G., Scheffler, B. E., Tellez, M. R., Rimando, A. M., Romagni, J. G., Duke, S. O., Nanayakkara, D., Khan, I. A., Abourashed, E. A., & Dayan, F. E. (2002). The inhibitory activity of natural products on plant *p*-hydroxyphenylpyruvate dioxygenase. *Phytochemistry*, *60*(3), 281–288. https://doi.org/10.1016/S0031-9422(02)00121-8

Meena, M., Prasad, V., & Upadhyay, R. S. (2016). Assessment of the bioweedicidal effects of *Alternaria alternata* metabolites against *Parthenium* species. *Bulletin of Environmental and Scientific Research*, *5*(1), 1–7. http://besr.org.in/index.php/besr/article/view/60

Meena, M., & Samal, S. (2019). *Alternaria* host-specific (HSTs) toxins: An overview of chemical characterization, target sites, regulation and their toxic effects. *Toxicology Reports*, *6*, 745–758. https://doi.org/10.1016/j.toxrep.2019.06.021

Moura, M. S., Lacerda, J. W. F., Siqueira, K. A., Bellete, B. S., Sousa, P. T., Óglio, E. L. D., Soares, M. A., Vieira, L. C. C., & Sampaio, O. M. (2020). Endophytic fungal extracts: Evaluation as photosynthesis and weed growth inhibitors. *Journal of Environmental Science and Health*, *55*(5), 470–476. https://doi.org/10.1080/03601234.2020.1721981

Nehl, D. B., Allen, S. J., & Brown, J. F. (1997). Deleterious rhizosphere bacteria: An integrating perspective. *Applied Soil Ecology*, *5*(1), 1–20. https://doi.org/10.1016/S0929-1393(96)00124-2

Pfirter, H. A., Ammon, H.-U., Guntli, D., Greaves, M. P., & Defago, G. (1997). Towards the management of field bindweed (*Convolvulus arvensis*) and hedge bindweed (*Calystegia sepium*) with fungal pathogens and cover crops. *Integrated Pest Management Reviews*, *2*(2), 61–69. https://doi.org/10.1023/A:1018432513776

Qiang, S., Zhu, Y., Summerell, B. A., & Li, Y. (2006). Mycelium of *Alternaria alternata* as a potential biological control agent for *Eupatorium adenophorum*. *Biocontrol Science and Technology*, *16*(7), 653–668. https://doi.org/10.1080/09583150600699804

Razaghi, P., & Zafari, D. (2017). *Phoma crystallifera* with phytotoxic effects and pathogenic potential against field bindweed (*Convolvulus arvensis* L.) in Iran. *Journal of Applied Microbiology*, *122*, 1275–1285. https://doi.org/10.1111/jam.13411

Reichert Júnior, F. W., Scariot, M. A., Forte, C. T., Pandolfi, L., Dil, J. M., Weirich, S., Carezia, C., Mulinari, J., Mazutti, M. A., Fongaro, G., Galon, L., Treichel, H., & Mossi, A. J. (2019). New perspectives for weeds control using autochthonous fungi with selective bioherbicide potential. *Heliyon*, *5*(5), e01676. https://doi.org/10.1016/j.heliyon.2019.e01676

Salvatore, M. M., Félix, C., Lima, F., Ferreira, V., Naviglio, D., Salvatore, F., Duarte, A. S., Alves, A., Andolfi, A., & Esteves, A. C. (2020). Secondary metabolites produced by *Macrophomina phaseolina* isolated from *Eucalyptus globulus*. *Agriculture*, *10*(3), 72. https://doi.org/10.3390/agricultura10030072

Saxena, S., & Kumar, M. (2007). Mycoherbicidal potential of *Alternaria alternata* ITCC4896 for the control of *Parthenium hysterophorus*. *Journal of Plant Protection Research*, *47*(2), 213–218.

Saxena, S., & Kumar, M. (2010). Evaluation of *Alternaria alternata* ITCC4896 for use as mycoherbicide to control *Parthenium hysterophorus*. *Archives of Phytopathology and Plant Protection*, *43*(12), 1160–1164. https://doi.org/10.1080/03235400802343825

Siddiqui, I., Bajwa, R., & Javaid, D. A. (2010). Mycoherbicidal potential of *Alternaria alternata* for management of *Chenopodium album* under field condition. *African Journal of Biotechnology*, *9*, 8308–8312. https://doi.org/10.5897/AJB10.1023

Sindhu, S. S., Khandelwal, A., Phour, M., & Sehrawat, A. (2018). Bioherbicidal potential of rhizosphere microorganisms for eco-friendly weed management. In V. S. Meena (Ed.), *Role of rhizospheric microbes in soil* (pp. 331–376). Springer. https://doi.org/10.1007/978-981-10-8402-7_13

Souza, A. R. C. D., Baldoni, D. B., Lima, J., Porto, V., Marcuz, C., Machado, C., Ferraz, R. C., Kuhn, R. C., Jacques, R. J. S., Guedes, J. V. C., & Mazutti, M. A. (2017). Selection, isolation, and identification of fungi for bioherbicide production. *Brazilian Journal of Microbiology*, *48*, 101–108. https://doi.org/10.1016/j.bjm.2016.09.004

Timmons, F. L. (1949). Duration of viability of bindweed seed under field conditions and experimental results in the control of bindweed seedlings. *Agronomy Journal*, *41*(3), 130–133. https://doi.org/10.2134/agronj1949.00021962004100030008x

Townsend, G., & Heuberger, J. (1943). Methods for estimating losses caused by diseases in fungicidal experiments. *Plant Disease Reporter*, *27*(17), 340–343. https://eurekamag.com/research/025/008/025008582.php

Tsuge, T., Harimoto, Y., Akimitsu, K., Ohtani, K., Kodama, M., Akagi, Y., Egusa, M., Yamamoto, M., & Otani, H. (2013). Host-selective toxins produced by the plant pathogenic fungus *Alternaria alternata*. *FEMS Microbiology Reviews*, *37*(1), 44–66. https://doi.org/10.1111/j.1574-6976.2012.00350.x

Tylkowska, K., Grabarkiewicz-Szczesna, J., & Iwanowska, H. (2003). Production of toxins by *Alternaria alternata* and *A. radicina* and their effects on germination of carrot seeds. *Seed Science and Technology*, *31*, 309–316. https://doi.org/10.15258/sst.2003.31.2.08

Verdugo-Navarrete, C., Maldonado-Mendoza, I. E., Castro-Martínez, C., Leyva-Madrigal, K. Y., & Martínez-Álvarez, J. C. (2021). Selection of rhizobacteria isolates with bioherbicide potential against Palmer amaranth (*Amarathus palmeri* S. Wats.). *Brazilian Journal of Microbiology*, *52*(3), 1443–1450. https://doi.org/10.1007/s42770-021-00514-2

Wan, Z. X., Zhu, J. J., & Qiang, S. (2001). The pathogenic mechanism of toxin of *Alternaria alternata* (Fr.) Keissler to *Eupatorium adenophorum*. *Journal of Plant Resources and Environment*, *10*, 47–50.

White, T. J., Bruns, T., Lee, S., & Taylor, J. (1990). Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In M. A. Innis, D. H. Gelfand, J. J. Sninsky, & T. J. White (Eds.), *PCR protocols* (pp. 315–322). Academic Press. https://doi.org/10.1016/B978-0-12-372180-8.50042-1

Zhang, X., Xi, H., Lin, K., Liu, Z., Yu, Y., Sun, Y., & Zhao, J. (2016). *Aspergillus* leaf spot of field bindweed (*Convolvulus arvensis* L.) caused by *Aspergillus niger* in China. *Springerplus*, *5*, 605. https://doi.org/10.1186/s40064-016-2292-4

Zhou, B., Wang, H., Meng, B., Wei, R., Wang, L., An, C., Chen, S., Yang, C., & Qiang, S. (2019). An evaluation of tenuazonic acid, a potential biobased herbicide in cotton. *Pest Management Science*, *75*(9), 2482–2489. https://doi.org/10.1002/ps.5402

Zonno, M. C., & Vurro, M. (1999). Effect of fungal toxins on germination of *Striga hermonthica* seeds. *Weed Research*, *39*(1), 15–20. https://doi.org/10.1046/j.1365-3180.1999.00119.x

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