


RESEARCH

Open Access



Morphological characterization, fungicidal alternatives and biological control of *Peronospora farinosa* on chamomile

M. M. Mergawy¹, M. M. H. Hassanin², A. A. M. Ali^{3*}  and Heba Yousef²

Abstract

Background Chamomile (*Matricaria chamomile* L.) is one of the very important medicinal plants worldwide, particularly in Egypt. The downy mildew disease is considered one of the most important diseases of chamomile. *Peronospora farinosa* was identified as the causal pathogen of downy mildew of infected chamomile plants, collected from Fayoum Governorate, Egypt, during 2022.

Results Efficiency of some fungicidal alternatives for management of downy mildew disease on chamomile leaves was evaluated in vitro studies using detached leaves technique. Each of the biocide, Bio-Cure F and Thyme oil emulsion treatments, before and after the fungal inoculation, respectively, was the best effective treatments, which reduced disease incidence to the lowest percentages. However, the treatment of Bio-Cure F before the fungal inoculation had the superiority for reducing disease severity to the lowest percentage, followed by Thyme oil emulsion, after the fungal inoculation. On the other hand, under greenhouse conditions, the best treatment reduced disease incidence and disease severity to the lowest percentages was Thyme oil emulsion, followed by Bio-Cure F, after and before the fungal inoculation, respectively. Total sugars content was obviously lower in treated chamomile plants before and after the fungal inoculation than in untreated controls. On the contrary, free phenols were obviously higher in treated chamomile plants before and after the fungal inoculation than in untreated controls.

Conclusion The present study indicated the possibility of using Bio-Cure F (biocide) and Thyme essential oil emulsion as spraying treatment against *P. farinosa* on chamomile plants.

Keywords Chamomile, Downy mildew, *Peronospora farinosa*, Bio-Cure F, Thyme essential oil

Background

Chamomile (*Matricaria chamomile* L.) is one of the most important and popular medicinal plants in Egypt. It belongs to family Asteraceae. In addition to being one

of the oldest and most commonly used medicinal plants, it is also well documented (Astin et al. 2000). It has been used for centuries as an antioxidant, astringent, curing medicine and an anti-inflammatory (Weiss 1988). Downy mildew disease is considered one of the most important diseases of chamomile in different cultivated areas. It was detected in Egypt for the first time in 1994, and *Peronospora sparsa* Berk. was identified as its causal pathogen (Hilal and Abd El-Moity 1994). Mergawy (2016) reported *Peronospora radii* (De Bary) that caused downy mildew of chamomile plants in Egypt.

Using chemical control against downy mildew disease leads to rejection of the shipment, due to the presence of chemical toxic residues in chamomile products.

*Correspondence:

A. A. M. Ali
abdallahali@agr.aswu.edu.eg

¹ Central Laboratory of Organic Agriculture, Agricultural Research Centre, Giza, Egypt

² Plant Pathology Research Institute, Agricultural Research Centre, Giza, Egypt

³ Plant Pathology Department, Faculty of Agriculture and Natural Resources, Aswan University, Aswan, Egypt

Trichoderma spp. are now considered the most common biological control agents against plant diseases due to their capabilities to inhibit phytopathogens and increase growth promotion in plants (Avşar et al. 2017). Thyme essential oil has an antifungal activity which might be due to its toxic effect (Šegvic Klaric et al. 2007) or due to its chitin penetration of fungal cell wall which damages the lipoprotein cytoplasmic membrane, leading to escape of cytoplasm (Zambonelli et al. 1996). The present investigation was carried out to evaluate some fungicidal alternatives for the management of downy mildew on chamomile that can overcome the risks of using fungicides, which are hazardous for humans and the environment.

Methods

Disease occurrence, isolation and identification of the causal pathogen of downy mildew on chamomile plants

Samples of chamomile plants infected with downy mildew were collected in January 2022 from a chamomile production fields in Abshaway and Yousef El-Seddik (Fayoum Governorate, Egypt). Symptoms included yellowing leaf and stem lesions with purple-gray sporulation, tip dieback and collapsed leaves. The disease survey was conducted during January and February 2022, and disease incidence was calculated based on the percentage of diseased plants in different areas of the field. Isolation of the pathogen was carried out using a pre-moistened leaf pieces covered with conidophores. Isolates were then maintained and multiplied on seedlings of chamomile plants, where re-inoculation is needed every 10–12 days. The pathogen was identified by its morphological and microscopic characteristics according to the method of Danielsen et al. (2002), and the identification was confirmed by the staff members of Department of Mycology and Plant Disease Survey, Plant Pathology Research Institute, Agricultural Research Center (ARC), Giza, Egypt. Ten leaf samples were examined via light microscopy, and 10 slides were prepared for each sample. Using a sterile microscopic needle, fungal structures were transferred from the plant surface to water drops to examine the presence of sporulation. The morphology of conidophores (appearance and branching pattern) and conidia was then also determined. Conidial germination and appressoria formation were examined on naturally infected plants after incubation for 24 h in moist conditions (in plastic bags) at 15 °C in the dark. After incubation, plant leaves were ripped apart and placed in water droplets on microscope slides and then examined via light microscopy.

Inoculum preparation

Downy mildew inoculum was prepared by collecting plant tissues having spots of the disease; then, they were placed in a baker contains sterilized water. Baker contains water and infected plant tissues were shaken on electric shaker for 20 min just to liberate fungal spores from tissues to liquid water. This suspension was used to inoculate healthy chamomile plants (Lebeda 1986). Conidial suspension was sprayed onto the healthy plants with atomizer sprayer (Lebeda 1984).

Evaluation of some alternative control methods efficacy against downy mildew on chamomile leaves in vitro

This experiment was carried out to evaluate the efficiency of Thyme essential oil (was equipped in Ornamental, Medicinal and Aromatic Plants Diseases Research Department, Plant Pathology Research Institute, ARC, Giza, Egypt), salicylic acid [(C₇H₆O₃), Sigma Company] and Bio-Cure F (*Trichoderma viride* 1 × 10⁶ cfu/g, M/S.T. Stanes Company Limit—India), as alternative control methods against downy mildew on chamomile leaves in laboratory and to find whether there is any correlation between obtained data from greenhouse and those obtained using detached leaves technique. The experiment was conducted in boxes (340 × 255 × 35 mm) lined with three layers of moistened cellulose cotton wool and one layer of filter paper, and healthy leaves of chamomile were placed on moistened filter paper. Chamomile leaves were sprayed individually with Thyme essential oil emulsion at rate 2 ml/l (Hassanin et al. 2017), salicylic acid at rate 0.5 g/l (Halawa et al. 2018) and Bio-Cure F 1.15% WP at rate 6 g/l (Hassanin et al. 2020), 24 h after or before the fungal inoculation, as mentioned in inoculum preparation. Twenty-seven leaves were used for each treatment. Each treatment was replicated three times. Boxes without treatments were used as a control. Boxes with inoculated leaves (treated and non-treated) are incubated in a cultivation room at a night/day temperature of 15 °C. The boxes were covered with foil after inoculation for 24 h. The final assessment of infection was carried out usually on day 7 after inoculation. Percentage of plants infected with downy mildew was recorded as disease percentage using the following formula according to El-Helaly et al. (1970) and Ahmed (2013):

$$\% \text{ Disease incidence} = \frac{\text{Number of infected plants}}{\text{Total plant numbers}} \times 100$$

Disease severity was calculated using scale developed and equation developed and modified by Yan et al. (2002) and Parkunan et al. (2013) as follows:

0 = no disease symptoms;
 1 = 1–25% coverage of lesions/plant;
 2 = 26–50% coverage of lesions/plant;
 3 = 51–75% coverage of lesions/plant;
 4 = 76–100% coverage of lesions/plant or completely collapsed plant.

Percentage of disease severity was recorded according to the following equation:

$$\text{Disease severity (\%)} = \frac{\sum n \times c}{N \times C} \times 100$$

where n is number of infected leaves, c category number, N total number of examined leaves and C the highest category number of infection.

Greenhouse experiment

Effect of some alternative control methods on chamomile downy mildew under greenhouse conditions

In greenhouse experiment, plastic pots (25×50 cm) in diameter were used. Each pot contains about 6-kg sterilized sandy loam soil, and five replicates were used for each treatment. Six chamomile transplants (35 day old) were transplanted in each pot. Chamomile plants were sprayed by Thyme essential oil emulsion at rate of 2 ml/l, salicylic acid at rate of 0.5 g/l and Bio-Cure F 1.15% WP at rate of 6 g/l, 24 h after or before the fungal inoculation, as mentioned in inoculum preparation. Percentage of plants infected with downy mildew was recorded, one week after each spray to determine percentage of disease incidence and disease severity using formula as mentioned before.

Biochemical changes associated with downy mildew infection

Samples of chamomile plants, treated with different alternatives, were collected from different replicates and mixed thoroughly, and then, one sample for each treatment was used to determine sugars content and phenols as follows:

Determination of sugars content

Total sugars were determined spectrophotometrically in chamomile samples using picric acid method described by Thomas and Dutcher (1924). Sugar content was expressed as mg glucose per one gram dry weight.

Determination of free, conjugated and total phenols

Samples of chamomile plants were used to determine: free, conjugated and total phenols using method described by Bray and Thorpe (1954).

Statistical analysis

The layout of this experiment was designed as factorial experiment in a complete randomized design with three replicates (Snedecor and Cochran 1989). This statistical analysis was done by using the computer program MS-TATEC software version (4) using the L.S.D. test at 0.05.

Results

Disease occurrence

Mean percentages of infected chamomile plants with downy mildew in Fayoum Governorate in January 2022 ranged between 7.6 and 8.1% in Yousef El-Seddik and Abshaway, respectively (av. 7.9%), whereas it gradually increased in February from 15.3 to 16.7% in Yousef El-Seddik and Abshaway, respectively (av. 16.0%).

Disease symptoms

Symptoms of the disease (Fig. 1) were firstly noticed on newly produced leaves of young plants as green spots, quickly turned yellow and then brown, died and became brittle. Infected young shoots may also become dry, wilted and die, slowing down the plant's growth. Under favorable conditions such as wetness of plant surfaces or high relative humidity, formed conidiophores of the pathogen were visible on mildew spots.

Identification of the causal pathogen of downy mildew

The causal pathogen was identified on chamomile plants as *Peronospora farinosa* via light microscopy according to the shape and branching ends of conidiophores and measurements of conidiophores and conidia. Microscopic observations showed the conidiophore on the



Fig. 1 Characteristic downy mildew symptoms on leaf surface, showing felt of conidiophores and conidia. **a** Conidiophores and conidia appear only on dead leaves. **b** Conidiophores and conidia appear in whole plant

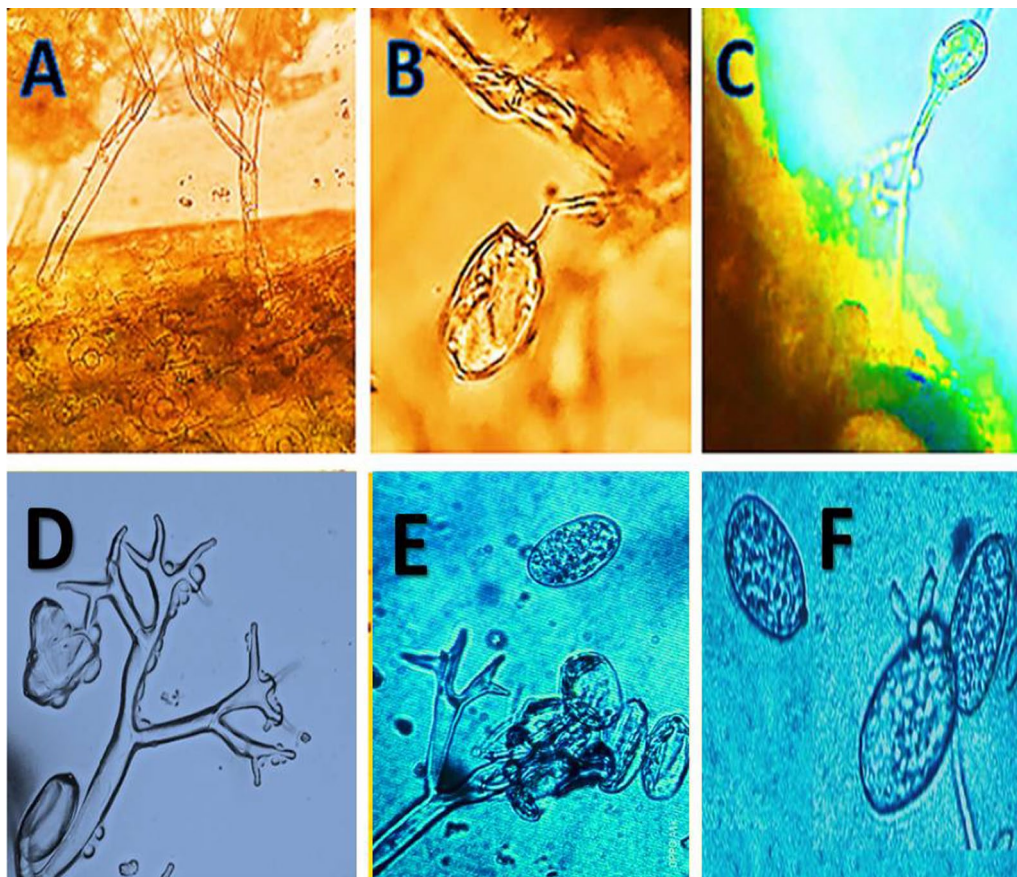


Fig. 2 Morphology of *Peronospora farinosa*. **a** Conidiophore on the surface of leaf. **b** Germinated conidium with appressorium formed on the surface of leaf. **c** Germination of conidia in a germ tube ending with appressoria. **d, e** Conidiophores which branched mainly dichotomously. **f** Sporangia

surface of leaf, which branched mainly dichotomously (200 to 400 μm long), four to five times with acutely tapering termini (Fig. 2a, d, e). Germinated conidium with appressorium was formed on the surface of leaves (Fig. 2b). Germination of conidia is in a germ tube ending with appressoria (Fig. 2c). Sporangia were ellipsoidal, light brown (Fig. 2f) and measured with mean dimensions of $22 \times 18 \mu\text{m}$.

Effect of some alternative control methods on chamomile downy mildew in vitro

In this experiment, chamomile leaves were sprayed individually with Thyme essential oil emulsion, salicylic acid and Bio-Cure F 1.15% WP, 24 h after or before the fungal inoculation to suppress the downy mildew disease. The disease incidence and severity were significantly reduced by the treatments compared with the untreated control.

Table 1 Effect of various control measures on disease incidence and severity of chamomile downy mildew before or after 24 h of fungal inoculation in vitro

Treatment	%Disease incidence		%Disease severity	
	Before fungal inoculation	After fungal inoculation	Before fungal inoculation	After fungal inoculation
Thyme oil emulsion	29.6	25.9	9.6	5.2
Salicylic acid	74.1	74.1	39.3	39.3
Bio-Cure F	25.9	51.9	4.4	12.6
Control	85.2	92.6	91.8	92.6
L.S.D.at 5%	4.6	2.5	4.9	5.7

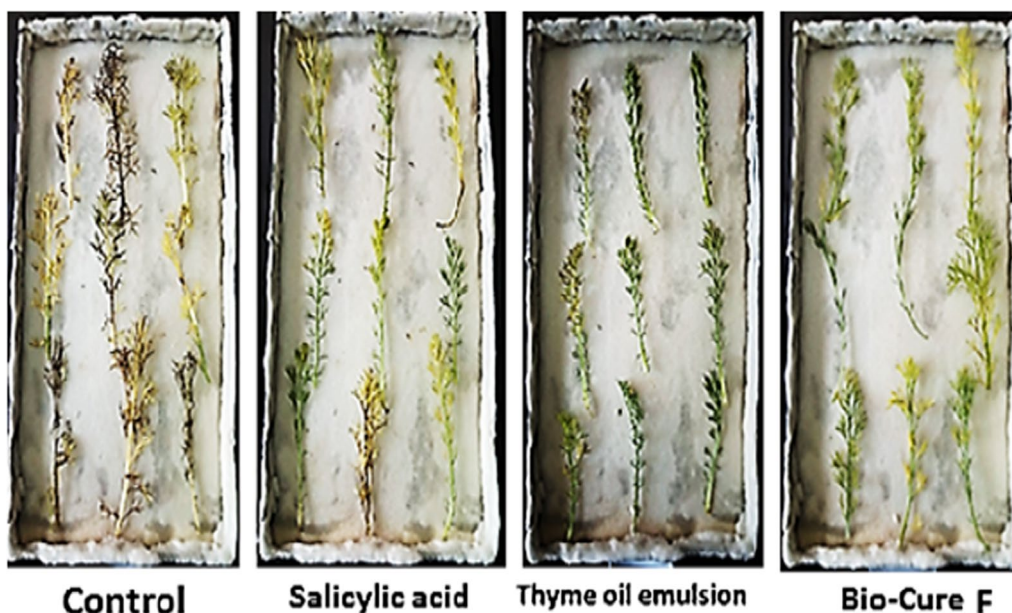


Fig. 3 Effect of spraying with treatments before inoculation with *Peronospora farinosa* (24 h)

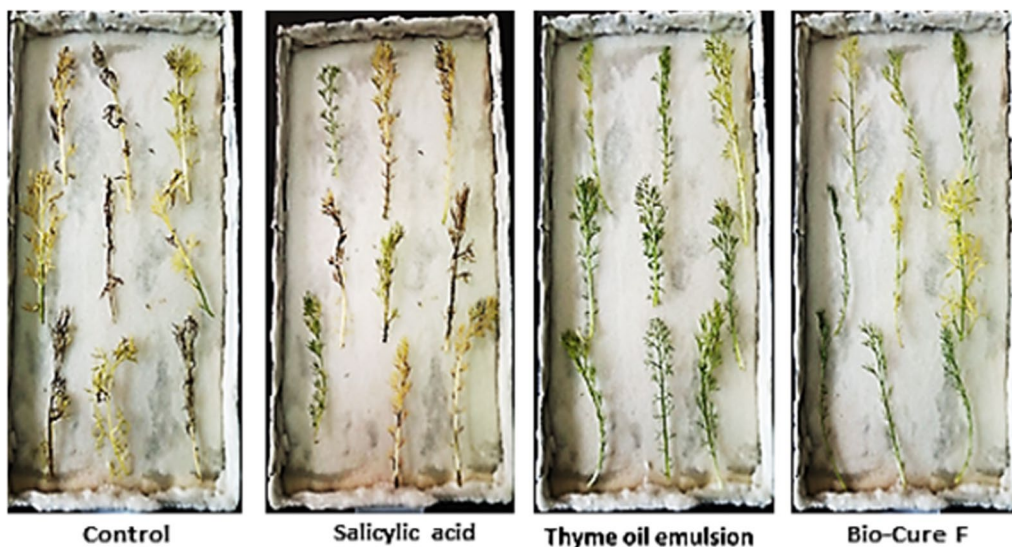


Fig. 4 Effect of spraying with treatments after inoculation with *Peronospora farinosa* (24 h)

Data presented in Table 1 and Figs. 3 and 4 showed that the lowest percentages of disease incidence (25.9%) were recorded in the treatments of Bio-Cure F and Thyme oil emulsion, before and after the fungal inoculation, respectively. However, the treatment of Bio-Cure F before the fungal inoculation, reduced disease severity to the lowest percentage (4.4%), followed by Thyme oil emulsion (5.2%) after the fungal inoculation.

Effect of some alternative control methods on chamomile downy mildew under greenhouse conditions

Data presented in Table 2 showed that the lowest disease incidence percentages were recorded in the treatments of Thyme oil emulsion (13.3%) and Bio-Cure F (19.9%), after and before the fungal inoculation, respectively, whereas the percentages of disease severity were the lowest in the treatment of Thyme oil emulsion

Table 2 Effect of various control measures on disease incidence and severity of chamomile downy mildew before or after 24 h of fungal inoculation under greenhouse conditions

Treatment	%Disease incidence		%Disease severity	
	Before fungal inoculation	After fungal inoculation	Before fungal inoculation	After fungal inoculation
Thyme oil emulsion	36.7	13.3	23.3	9.7
Salicylic acid	26.7	37.5	19.7	35.6
Bio-Cure F	19.9	43.3	12.8	29.4
Control	86.7	83.3	59.2	51.7
L.S.D.at 5%	4.1	2.7	4.6	5.0

(9.7%) and Bio-Cure F (12.8%), after and before the fungal inoculation, respectively.

Effect of treatment with fungicidal alternatives on total sugar content, free, conjugated and total phenols of chamomile plants

Data in Table 3 show that total sugars content was obviously lower in treated chamomile plants before and after the fungal inoculation than in untreated controls. The results of treatments application before the fungal inoculation showed that the maximum values of total sugars were recorded in control treatment (29.20 mg/g dry weight), followed by Thyme oil emulsion (13.76 mg). The intermediate values were resulted by salicylic acid (12.56 mg). Meanwhile, the lowest values of total sugars were recorded in plants treated with Bio-Cure F (6.98 mg). On the other hand, the results of treatments application after the fungal inoculation showed that the maximum values of total sugars were recorded in control treatment (32.55 mg/g dry weight), followed by salicylic acid (16.06 mg). The intermediate values were resulted by Bio-Cure F (13.83 mg). Meanwhile, the lowest values of total sugars were recorded in plants treated with Thyme oil emulsion (9.38 mg).

Data in Table 3 showed also that the free phenols were obviously increased in treated chamomile plants before and after the fungal inoculation than in untreated

controls. The maximum values of free phenols, in treatments applied before the fungal inoculation, were recorded in Bio-Cure F treatment (66.10 mg/g dry weight), followed by salicylic acid (62.63 mg). On the other hand, the maximum values of free phenols, in treatments applied after the fungal inoculation, were recorded in Thyme oil emulsion treatment (74.83 mg/g dry weight), followed by Bio-Cure F (61.46 mg). Conjugated and total phenols were obviously increased in treated chamomile plants before and after the fungal inoculation than in untreated controls. The maximum values of conjugated and total phenols, 41.26 and 103.89 mg/g dry weight, respectively, in treatments applied before the fungal inoculation, were recorded in plants treated with salicylic acid treatment. On the other hand, the maximum values of conjugated and total phenols, 56.20 and 111.36 mg/g dry weight, respectively, in treatments applied after the fungal inoculation, were recorded in salicylic acid treatment.

Discussion

In the present research, samples of chamomile plants infected with downy mildew were collected from a chamomile production fields in Fayoum Governorate, Egypt. The symptoms of the infected plants sound a lot like the ones described by Mergawy (2016). However, according to method of Danielsen et al. (2002), the causal fungus

Table 3 Effect of various control measures of chamomile downy mildew on total sugars content, free, conjugated and total phenols (mg/g dry weight) before or after 24 h of fungal inoculation under greenhouse conditions

Treatment	Total sugars content		Before fungal inoculation			After fungal inoculation		
	Before fungal inoculation	After fungal inoculation	Phenols			Phenols		
			Free	Conjugated	Total	Free	Conjugated	Total
Thyme oil emulsion	13.76	9.38	55.53	29.50	85.03	74.83	12.03	86.86
Salicylic acid	12.56	16.06	62.63	41.26	103.89	55.16	56.20	111.36
Bio-Cure F	6.98	13.83	66.10	28.16	94.26	61.46	30.40	91.86
Control	29.20	32.55	20.96	24.48	45.44	22.83	72.76	95.59

of downy mildew was identified as *Peronospora farinosa*, which may be isolated for the first time from chamomile plants in Egypt. The global climate changes play an important role and impact on the appearance and distributions of pathogens. The degrees and types of the effect differed according to the geographical situation of the respective countries. Climate changes had a significant impact on the appearance and intensity of infection with downy mildew on chamomile, since the low temperatures with high humidity led to the emergence of severe infection.

Fungicides are the most important method of controlling the plant diseases. The use of fungicides, however, is harming to humans, the environment and living organisms (Guzzo et al. 1993). Abiotic agents are considered alternative control methods to fungicides for giving plants acquired resistance against pathogens. In the present study, some alternative control methods were evaluated for management of downy mildew disease. In vitro studies, chamomile leaves were sprayed with Thyme essential oil emulsion, salicylic acid and Bio-Cure F 1.15% WP, 24 h after or before the fungal inoculation. Treatments with Bio-Cure F and Thyme oil emulsion, before and after fungal inoculation, respectively, were the most effective and reduced disease incidence to the lowest percentages. Nevertheless, Bio-Cure F before the fungal inoculation was the most effective at reducing disease severity, followed by Thyme oil emulsion after the fungal inoculation. The positive effect of Bio-Cure F before fungal infection may play two roles as: (1) defense against pathogenic infection, when the fungal conidia germinate and penetrate the host by the germ tube with appressoria and (2) as inducer to the leaves to produce phytoalexins which forbidden the plant infection. These data are in agreement with those obtained by Mahmoud et al. (2006) who reported that induced resistance was associated with increased activity and accumulation of oxidative enzymes (peroxidase and polyphenoloxidase) in root tissues. Also, biological control using microorganisms or their secretions prevented or reduced plant diseases. These results were also, to somewhat, similar to those reported by Naglot et al. (2015) who reported that *Trichoderma viride* exhibited significant antifungal activity against five common phytopathogenic fungi. According to authors, the cultural filtrate of the antagonist from stationary growth phase exhibited significantly more inhibitory activity against all of the tested fungi, indicating that extracellular antifungal metabolites were present at optimal concentrations. Furthermore, the role of the enzymes in cultural filtrates was examined quantitatively and it was determined that pectinase, cellulase, β -1, 3-glucanase and amylase

activities were the highest in the exponential phase, while proteases and chitinase activity were the highest in the stationary phase. On the other hand, the positive effect of Thyme oil emulsion after fungal infection may be due to anti-fungal effect, because of the presence of active substances in essential oils such as thymol (Hassanin et al. 2017). Many scientists have interpreted the mode of action of active substances in the essential oils of medicinal and aromatic plants (Hassanin et al. 2017).

Under greenhouse conditions, Thyme oil emulsion was the most effective treatment in reducing disease incidence and severity, followed by Bio-Cure F. The efficacy of the biocide, Bio-Cure F, on chamomile plant may be due to inducing resistance. The reduction in the disease incidence as a result of using tested biocide might be attributed to produce some growth regulators that stimulate plant to tolerate infection or working as resistance producers (De Curtis et al. 2010). *Trichoderma* spp. have some mechanisms of action on fungal pathogens (Harman 2006), production of some destructive enzymes such as cellulase and chitinase that degrade pathogen cell wall (Bolar et al. 2000). On the other hand, the effect of an emulsion of Thyme oil against downy mildew on chamomile may be due to the antifungal effect of some active substances presented in the oil. According to Šegvic Klaric et al. (2007), thymol, 1, 8-cineole and p-cymene in thyme essential oil are responsible for its antifungal activity. Hassanin et al. (2017) found that Thyme essential oil emulsion had the best antifungal effect at the lowest concentrations.

Total sugars content was clearly lower in chamomile plants treated before and after fungal inoculation compared to untreated controls. The highest soluble sugar indicated that untreated plants may have more efficient sugar synthesizing capacity which might be supporting the mycelium growth and sporulation. Moreover, availability of low sugar content in the intracellular fluid of the treated plants may be responsible for the inhibition of the growth and multiplication of the pathogen (Mahatma et al. 2009).

In contrast, free phenols were evidently higher in chamomile plants treated before and after fungal inoculation than untreated controls. The presence of hydroxyl groups in phenols makes them good electron donors, which can contribute to antioxidant effects; free phenols are responsible for inducing plant resistance (Mergawy 2016). Phenolic compounds play an important role in protective agents, inhibitors and fungicides against fungal pathogens (Bhattacharya et al. 2010). Also, there is an evidence that phenolic compounds suppress free radical production in biological systems and scavenge oxygen (Aryal et al. 2019). Furthermore, *Trichoderma* spp. spores enhanced phenolic compounds in healthy plants, although this improvement was less than infected plants.

Conclusion

Peronospora farinose was identified as the causal pathogen of downy mildew of infected chamomile plants collected from Fayoum Governorate, Egypt, during 2022 season, using the morphological identification. Some fungicidal alternatives, Thyme essential oil, salicylic acid and Bio-Cure F (*Trichoderma viride*), were evaluated for their efficacy against downy mildew of chamomile. Bio-Cure F and Thyme essential oil emulsion were the best treatments for the management of chamomile downy mildew, which indicated the possibility of their using as fungicidal alternatives.

Acknowledgements

Not applicable.

Author contributions

All contributions for the whole article belong to the authors. All authors read and approved the final manuscript.

Funding

Not applicable.

Availability of data and materials

The data and materials of this study have been presented in the manuscript.

Declarations

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

Received: 11 May 2023 Accepted: 14 June 2023

Published online: 03 July 2023

References

- Ahmed MFA (2013) Studies on non-chemical methods to control some soil borne fungal diseases of bean plants *Phaseolus vulgaris* L. Ph.D. thesis, Faculty of Agriculture Cairo University (Egypt)
- Aryal S, Baniya MK, Danekhu K, Kunwar P, Gurung R, Koirala N (2019) Total phenolic content, flavonoid content and antioxidant potential of wild vegetables from Western Nepal. *Plants* 8:96. <https://doi.org/10.3390/plants8040096>
- Astin JA, Pelletier KR, Marie A, Haskell WL (2000) Complementary and alternative medicine use among elderly persons: One-year analysis of a blue shield medicare supplement. *J Gerontol Med Sci* 55:4–9
- Avşar C, Koyuncu H, Aras ES (2017) Isolation and molecular characterization of *Bacillus* spp. isolated from soil for production of industrial enzymes. *Biol Chem Res* 3:72–86
- Bhattacharya A, Sood P, Citovsky V (2010) The roles of plant phenolics in defense and communication during Agrobacterium and Rhizobium infection. *Mol Plant Pathol* 11:705–719. <https://doi.org/10.1111/j.1364-3703.2010.00625.x>
- Bolar JP, Norelli JL, Wong KW, Hayes CK, Harman QE, Aldwinckle HS (2000) Expression of endochitinase from *Trichoderma harzianum* in transgenic apple increases resistance to apple scab and reduces vigor. *Phytopathology* 90:72–77
- Bray HG, Thorpe WW (1954) Analysis of phenolic compounds of interest in metabolism. *Meth Biochem Anal* 1:27–52
- Danielsen S, Jacobsen SE, Hockenhull J (2002) First report of downy mildew of quinoa caused by *Peronospora farinosa* f. sp. *chenopodii* in Denmark. *Plant Dis* 86:1175. <https://doi.org/10.1094/PDIS.2002.86.10.1175B>
- De Curtis F, Lima G, Vitullo D, De Cicco V (2010) Biocontrol of *Rhizoctonia solani* and *Sclerotium rolfsii* on tomato by delivering antagonistic bacteria through a drip irrigation system. *Crop Prot* 29:663–670
- El-Helaly AF, Elarosi HM, Assawah MW, Abol-Wafa MT (1970) Studies on damping-off and root rots of bean in UAR (Egypt). *United Arab Rep J Phytopathol* 2:41–57
- Guzzo SD, Bach EE, Martins EF, Moraes WC (1993) Crude exopolysaccharides (EPS) from *Xanthomonas campestris* pv. *manihotis*, *Xanthomonas campestris* pv. *campestris* and commercial xanthan gum as inducers of protection in coffee plants against *Hemileia vastatrix*. *J Phytopathol* 139:119–128
- Halawa AEA, Ali AAM, Hassanin MMH (2018) Efficiency of some organic acids as safe control mean against root and stem rot disease of *Coleus forskohlii*. *J Phytopathol Pest Manag* 5:48–62
- Harman GE (2006) Overview of mechanisms and uses of *Trichoderma* spp. *Phytopathology* 96:190–194
- Hassanin MMH, Abd-El-Sayed MA, Abdallah MA (2017) Antifungal activity of some essential oil emulsions and nanoemulsions against *Fusarium oxysporum* pathogen affecting cumin and geranium plants. *Sci J Flowers Ornament Plants* 4:245–258
- Hassanin MMH, Nada MGA, Halawa AEA (2020) Evaluation efficacy of some agents as safe alternatives to fungicides in management root-rot and wilt diseases of *Artemisia absinthium* and *A. santonicum*. *Egypt J Phytopathol* 48:94–106
- Hilal AA, Abd El-Moity TH (1994) Recent outbreaks of downy mildews on chamomile and rose plants. *First Conf Ornamental Hort* 2:638–651
- Lebeda A (1984) Response of differential cultivars of *Lactuca sativa* to *Bremia lactucae* isolates from *Lactuca serriola*. *Trans Br Mycol Soc* 83:491–494
- Lebeda A (1986) *Bremia lactucae*. In: Lebeda A (ed) *Methods of testing vegetable crops for resistance to plant pathogens*. VJH Sempra, Res Breed Inst Veget Crops, Olomouc, pp 105–113
- Mahatma MK, Bhatnagar R, Dhandhukia P, Thakkar VR (2009) Variation in metabolites constituent in leaves of downy mildew resistant and susceptible genotypes of pearl millet. *Physiol Mol Biol Plants* 15:249–255
- Mahmoud EY, Shokry SYM, Hussin ZN (2006) Induction of resistance in peanut plants against root rot diseases under greenhouse conditions by some chemical inducers. *J Agric Sci Mansoura Univ* 31:3511–3524
- Mergawy MM (2016) Control of foliage diseases in chamomile (*Matricaria chamomile* L.) plants under organic agriculture regulations. Ph. D. thesis, Faculty of Agriculture, Fayoum University
- Naglot A, Goswami S, Rahman I, Shrimali DD, Yadav KK, Gupta VK, Rabha AJ, Gogoi HK, Veer V (2015) Antagonistic potential of native *Trichoderma viride* strain against potent tea fungal pathogens in North East India. *Plant Pathol J* 31:278–289
- Parkunan V, Gitaitis RD, Dutta B, Langston DB, Ji P (2013) An epidemic of downy mildew caused by *Peronospora destructor* on *Vidalia* sweet onions in Georgia in 2012. *Plant Health Prog* 14:54. <https://doi.org/10.1094/php-2013-0328-01-br>
- Šegvić Klarić M, Kosalec I, Mastelić J, Piecková E, Pepeljnak S (2007) Antifungal activity of thyme (*Thymus vulgaris* L.) essential oil and thymol against moulds from damp dwellings. *Lett Appl Microbiol* 44:36–42
- Snedecor GW, Cochran WG (1989) *Statistical methods*, 8th edn. Iowa State University Press, Ames
- Thomas W, Dutcher RA (1924) The colorimetric determination of carbohydrates in plants by the picric acid reduction method. 1. The estimation of reducing sugars and sucrose. *J Am Chem Soc* 46:1662–1669
- Weiss RF (1988) In: Arcanum AB (ed) *Herbal medicine*. Beaconsfield Publishers, Beaconsfield, pp 22–28
- Yan Z, Reddy MS, Ryu CM, McInroy JA, Wilson M, Kloepper JW (2002) Induced systemic protection against tomato late blight elicited by plant growth-promoting rhizobacteria. *Phytopathology* 92:1329–1333
- Zambonelli A, d'Aulerio AZ, Bianchi A, Albasini A (1996) Effects of essential oils on phytopathogenic fungi *in vitro*. *J Phytopathol* 86:491–494

Publisher's Note

Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.