

ANTIFUNGAL ACTION OF DIMETHYL SULFOXIDE TOWARDS PLANT PATHOGENIC FUNGIES

Donyo Ganchev

Agricultural University, town of Plovdiv, Bulgaria, donyo@abv.bg

ORCID ID: 0000-0002-3776-7964

Abstract: *In vitro* tests for antifungal activity with dimethyl sulfoxide (DMSO) was conducted towards three widely spread and damaging plant parasitic fungus *Monilinia fructigena* causing brown rot of fruit on orchard plants mostly on cherries and sour cherries and other pome fruit cultures, *Erysiphe necator* (*Uncinula necator*) and *Blumeria graminis* (*Erysiphe graminis*) causing powdery mildews on grapes and wheat. All plant pathogens were from *Ascomycota* division and cause seriously damages annually on infected plant cultures. The *in vitro* tests were conducted by three different methods: Conidial Tests (Germ tube inhibition tests) for establishing the possibility of inhibition conidia spores of *Monilinia fructigena*; a radial growth mycelium assays for establishing mycelia growth inhibition of *Monilinia fructigena* and leaf disks *in vitro* tests for *Uncinula necator* and *Erysiphe graminis*. Results confirm the antifungal action of DMSO towards some plant pathogens particularly – towards *Monilinia fructigena*. The chemical was able fully to block the mycelium of *Monilinia fructigena* at 1 % concentration (v/v). Below this concentration, the action of DMSO rapidly decrease and at 0.5 % (v/v) concentration this chemical do not express and antifungal activity towards this plant pathogen in mycelium inhibition radial growth assays. However, in the conducted *in vitro* tests for inhibition of germination of conidia spores of *Monilinia fructigena*, DMSO even slightly stimulate spore germination, which strongly limited application of this compound as a cheap, low toxic fungicide against of *Monilinia fructigena*. DMSO does not express any antifungal activity against tested powdery mildews of *Uncinula necator* and *Erysiphe graminis* infested grapes and wheat. The same fungicidal action of DMSO was established towards *Botrytis cinerea*, *Fusarium graminearum* and *Aspergillus parasiticus* in the other studies. The present research confirm this. In all cases of established fungicidal action of DMSO, inhibited plant paratactic fungus from this compound was from division of *Ascomycota*. Although other research shows that DMSO can inhibit spores germination of *Tilletia laevis* and *Fusarium graminearum*, the present study shows that towards *Monilinia fructigena* was the opposite – DMSO even stimulate the germination of conidiospores

Keywords: dimethyl sulfoxide, DMSO, *Monilinia fructigena*, *Uncinula necator*, *Erysiphe graminis*,

1. INTRODUCTION

Dimethyl Sulfoxide is a cheap clear odorless liquid, produced as a by-product of the paper industry. This organic chemical is commonly used as dissolver in chemistry including different drugs and toxic compounds (Swanson, 1985). Dimethyl sulfoxide has many applications in the pharmacy as anti-inflammation, diuresis, analgesia action. The compound has excellent and rapid cell membrane penetration which is used for carry different drugs into target tissues / cells (Jacob and Wood, 1967). This chemical can very rapidly penetrate into skin approximately 5 minutes after cutaneous application (Manjunath and Shivaprakash, 2013). However the toxicity of DMSO is relatively low including towards humans, animals, plants and algae species (Brayton, 1986; Galvao et al., 2014; Ganchev, 2026). Other study found that DMSO treatment substantially altered the morphology and attachment of cells in concurrence with a significant reduction in cell viability in a dose-dependent manner (Pal et al., 2012). Conventional treatment of osteoarthritis with DMSO need to be applied with a caution due to the possible unbinding, and questionable treatment duration and dose (Brien et al., 2008). In the agriculture, dimethyl sulfoxide was found to express antifungal effect towards grey mold (*Botrytis cinerea*) with strong growth inhibition at 0.5 % concentration. However, 2 % water solutions of DMSO can cause severe injuries on tomato plants (Petrucelli et al., 2020). Was found that application of DMSO in concentrations of 0.01 to 0.1 % to the soil or as foliar sprays of 0.001 to 0.01 % can increase grain yields of rice plants (Kumar et al., 1976). DMSO in over 2.5 % concentrations can significantly reduce the germination of teliospores of *Tilletia laevis* teliospore germination in conducted *in vitro* tests (Ansari et al., 2015). Other study reveal that this compound can inhibit mycelial growth and conidial germination of *Fusarium graminearum* (Li et al., 2025). Dimethyl sulfoxide also was found to have antifungal effect on fungus which degraded archaeological wood as *Aspergillus parasiticus*. Even more DMSO had no effect on the color, structure and chemical characteristic of the wood (Osman et al., 2017). Other fungi which can be inhibit by DMSO is *Sclerotium rolfsii* (Melhuish and Bean, 1971). Traditionally DMSO is used in agriculture for chlorophyll extraction (Barnes et al., 1992; Shinano et al., 1996; Kumari et al., 2018) or of carotenoids (Dardani et al., 2024) and a starch (Snyder et al., 1977; Dintzis et al., 1995) or various other organic compounds (Burnouf and Bietz, 1989; Park and Martinez, 1992).

Dimethyl sulfoxide, soil-incorporated at rates of 20 to 240 gpa, prevented the growth of roots and delayed and inhibited the growth of shoots of plants developing from purple nutsedge (*Cyperus rotundus* L.) tubers for a

period of about two months was established in the conducted study prove herbicidal activity of this chemical (Anderson and Dunford, 1966). Four percent concentration of DMSO showed a significant effect in the reduction of themicrobial degradation in strawberries as post harvest treatments (Oliveira at al., 2018). Even insecticidal activity of Dimethyl sulfoxide towards *Drosophila melanogaster* was reported with LC50 = 0.42 % (Cvetković et al., 2015) and acaricidal activity of hatching of eggs inhibition of *Rhipicephalus (Boophilus) annulatus* ticks too (Ravindran at al., 2011). Antiparasitic action of DMSO towards *Nosema ceranae* - a microsporidian, unicellular parasite which affect honey bees was also found (Ho at al., 2024)

In the present research in vitro tests with three widely spread plant pathogens from division Ascomycota were conducted for investigation the possible fungicidal action of DMSO. This plant parasitic fungus were: *Monilinia fructigena* causing brown rot of fruit on orchard plants mostly on cherries and sour cherries and other pome fruit cultures (Van Leeuwen, 2000), *Erysiphe necator (Uncinula necator)* (Gadoury at al., 2012) and *Blumeria graminis (Erysiphe graminis)* (Graeff et al., 2006) casing powdery mildews on grapes and wheat.

2. MATERIALS AND METHODS

n vitro tests for antifungal activity were performed by three different methods:

Conidial Tests (Germ tube inhibition tests) for establishing the possibility of inhibition conidia spores of plant pathogens were carried out by spraying of microscopic slides pattern “hanging drop” with water solutions of DMSO in different concentrations. After drying the solution, a conidial suspension made from relevant phytopathogen (3*10⁴ spores / ml – 20 µl) was added. The slides were incubated in thermostat (humid chamber) under 22 – 24°C. Two days after, observations via light microscope 400 x magnification were conducted for evaluation of germination of conidia spores (Antonov et al., 1977; Wang and Higgins, 2005).

For establishing mycelia growth inhibition, a radial growth assays tests were conducted in 10 cm diameter Petri dishes with potato dextrose agar media (PDA). In sterile Petri 1 ml of water solution of DMSO in different concentration was added plus 9 ml of PDA. After cooling the mixture one 10 mm diameter inoculation PDA disk from relevant phytopathogen culture was placed in the center of Petri. Inoculated petri dishes was incubated under temperature interval of 22 – 25°C. The observations were made 7 and 14 days after inoculation – with was determinate a growth of the mycelium in mm around of the inoculated discs. The effectiveness was calculated by the formula of Abbot. (El-Masry, 2002; Taskeen-Un-Nisa at al., 2011; Piepho et al., 2024)

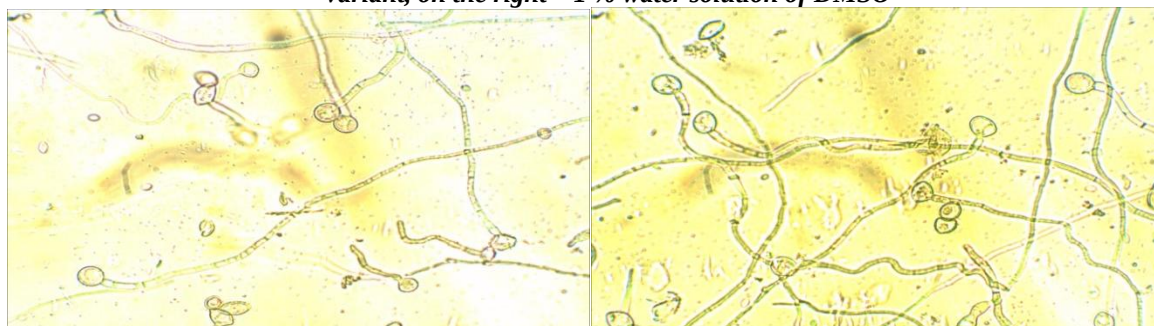
Leaf disks in vitro tests were conducted with obligatory plant pathogens which cannot be grown on the artificial media. The 10 mm disks from naturally infected leaves with the given pathogen were placed onto 6 cm wide and 20 cm long double filter paper placed onto 10 cm long and 6 cm wide polycarbonate sheets. polycarbonate sheets were placed onto 15 cm long and 10 cm wide plastic plates filled to the top with distilled water. The middle of the filter paper was on the polycarbonate sheet, while the ends were dropped into the water. The leaf disk were preliminarily soaked with distilled water solution of DMSO in different concentrations. Organosilicon surfactant was added at 0.1 % to this solutions for improving the wetting ability. The control variant was treated only with water solution of organosilicon surfactant in 0.1 % concentration (Peng and Kuc, 1992; Agnola et al., 2003).

In vivo trials for phytotoxicity were conducted with 3 varieties of cherry orchard tree, wheat and grapes. The plants were sprayed with distilled water solutions of DMSO in different concentrations. After 1 and 2 weeks visual observation of phytotoxicity (necrosis, chlorosis, whitening's, deformations) were performed

3. RESULTS

In the conducted conidial spores inhibition in vitro tests, dimethyl sulfoxide express even stimulation effect on germination of conidia in comparison with the control variant – more spores were germinated in the comparison with the control variant and they were with longer germs (Figure. 1).

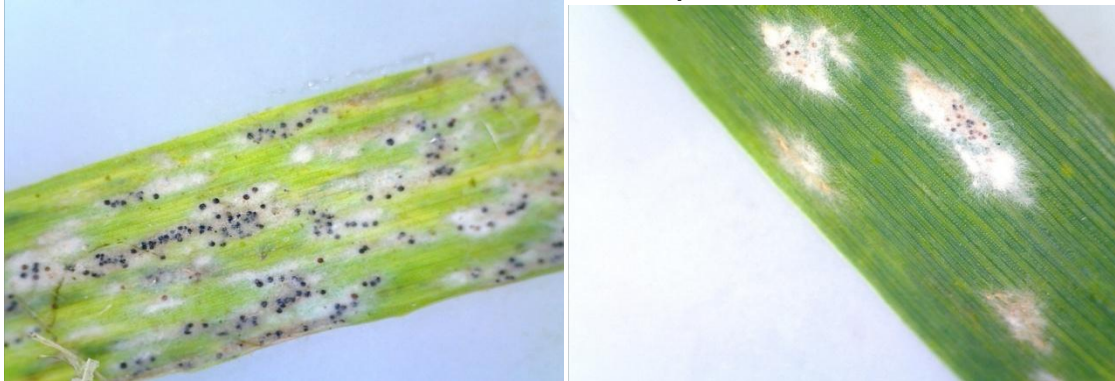
Figure 1. Effect of dimethyl sulfoxide towards conidia spores of *Monilinia fructigena*. On the left – control variant, on the right – 1 % water solution of DMSO



Source: Author's research

Below 1 % concentration, waster solutions of DMSO do not affect germination of conidia i.e. the effect was the same like the control variant. . However above this concentration, stimulation effect on germination of spores of this plant pathogen appeared. Increasing concentrations to 4 % increase this stimulation effect of DMSO. The conducted leaf disk tests with *Erysiphe graminis* shows that DMSO did not express any fungicidal activity towards this pathogen (Figure. 2)

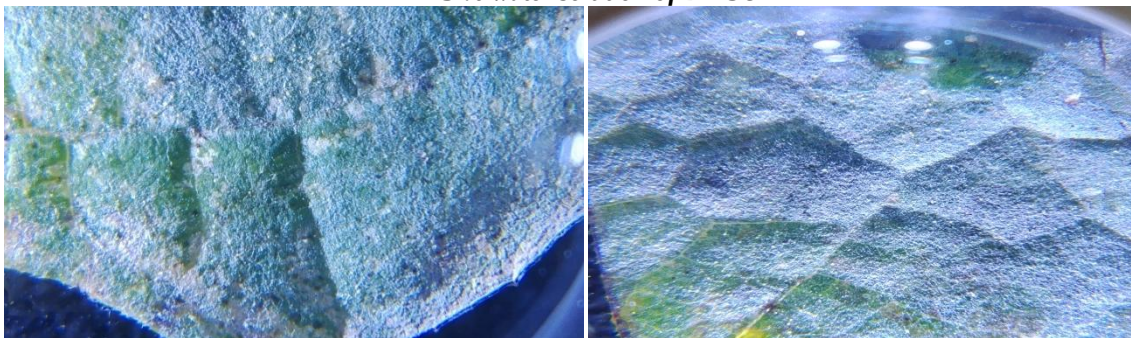
Figure 2. Effect of dimethyl sulfoxide towards *Erysiphe graminis* . On the left – control variant, on the right – 3 % water solution of DMSO



Source: Author's research

The same was found towards *Uromyces necator* – there is no difference with the control variant (Figure.3)

Figure 3. Effect of dimethyl sulfoxide towards *Uromyces necator*. On the left – control variant, on the right – 3 % water solution of DMSO



Source: Author's research

Figure 4. Effect of dimethyl sulfoxide towards mycelium of *Monilinia fructigena*. On the left – control variant, on the right – 1 % water solution of DMSO



Source: Author's research

However, in conducted in vitro test for mycelium inhibition in potato dextrose agar (PDA), DMSO was able fully to block development of mycelium of *Monilinia fructigena* at 1 % (v/v) concentration. Below this concentration, the action of DMSO rapidly decrease and at 0.5 % (v/v) concentration this chemical do not express antifungal activity towards this plant pathogen in mycelium inhibition radial growth assays. Conducted phytotoxicity tests reveal that up to 3-4 % concentration, water solutions of DMSO do not cause any damages on treated cherry, wheat and grape plants which means that DMSO can be safely sprayed onto plants.

4. DISCUSSIONS

The conducted in vitro tests, confirm the antifungal action of DMSO towards some plant pathogens particularly – towards *Monilinia fructigena* in this research. The same action was established towards *Botrytis cinerea* (Petrucelli et al., 2020), *Fusarium graminearum* (Li et al., 2025), *Aspergillus parasiticus* (Osman et al., 2017) and *Sclerotium rolfsii* (Melhuish and Bean, 1971). In all cases of established fungicidal action of DMSO, inhibited plant paratactic fungus from this compound was from division of Ascomycota. DMSO however was found to even stimulate the germination of conidia spores of *Monilinia fructigena* although that in the other research this chemical significantly reduce the germination of teliospores of *Tilletia laevis* (Ansari et al., 2015) and conidial germination of *Fusarium graminearum* (Li et al., 2025)

5. CONCLUSIONS

The presented study shows antifungal action of dimethyl sulfoxide (DMSO) towards mycelium of *Monilinia fructigena*. The compound was able fully to inhibit this plant pathogen in the conducted in vitro trials at 1 % (v/v) concentration in potato dextrose agar media (PDA). In the conducted phytotoxicity tests, DMSO do not injury the plants of cherry, wheat and grape even when was applied at 3-4 % (v/v) concentration. However, in the conducted in vitro tests for inhibition of germination of conidia spores of *Monilinia fructigena*, DMSO even slightly stimulate spore germination, which strongly limited application of this compound as a cheap, low toxic fungicide against of *Monilinia fructigena*. DMSO do not express any antifungal activity against tested powdery mildews of *Ucinula necator* and *Erysiphe graminis* infested grapes and wheat.

REFERENCES

- Agnola, B., Boury, S., Monot, C., Quillévéré, A., Hervé, Y., & Silué, D. (2003). Evidence that a leaf-disk test allows assessment of isolate-specific resistance in Brassica oleracea crops against downy mildew (*Peronospora parasitica*). *European Journal of Plant Pathology*, 109(5), 471–478. doi.org
- Anderson, W. P., & Dunford, M. P. (1966). The herbicidal activity of dimethyl sulfoxide on purple nutsedge. *Weeds*, 14(3), 195–197. doi.org
- Ansari, S., Moosavi, M. R., Carris, L. M., Nasrollahi, M., & Mirzaee, M. R. (2015). Dimethyl sulfoxide inhibits *Tilletia laevis* teliospore germination. *Australasian Mycologist*, 32, 2–5.
- Antonov, A., Stewart, A., & Walter, M. (1997, August). Inhibition of conidium germination and mycelial growth of *Botrytis cinerea* by natural products. In *Proceedings of the New Zealand Plant Protection Conference* (Vol. 50, pp. 159–164).
- Barnes, J. D., Balaguer, L., Manrique, E., Elvira, S., & Davison, A. W. (1992). A reappraisal of the use of DMSO for the extraction and determination of chlorophylls a and b in lichens and higher plants. *Environmental and Experimental Botany*, 32(2), 85–100. doi.org
- Brayton, C. F. (1986). Dimethyl sulfoxide (DMSO): A review. *The Cornell Veterinarian*, 76(1), 61–90.
- Brien, S., Prescott, P., Bashir, N., Lewith, H., & Lewith, G. (2008). Systematic review of the nutritional supplements dimethyl sulfoxide (DMSO) and methylsulfonylmethane (MSM) in the treatment of osteoarthritis. *Osteoarthritis and Cartilage*, 16(11), 1277–1288. doi.org
- Burnouf, T., & Bietz, J. A. (1989). Rapid purification of wheat glutenin for reversed-phase high-performance liquid chromatography: Comparison of dimethyl sulfoxide with traditional solvents. *Cereal Chemistry*, 66(2), 121–127.
- Cvetković, V. J., Mitrović, T. L., Jovanović, B., Stamenković, S. S., Todorović, M., Đorđević, M., & Radulović, N. (2015). Toxicity of dimethyl sulfoxide against *Drosophila melanogaster*. *Biologia Nyssana*, 6(2), 91–95.
- Dardani, C. S., Machado, W. R. C., Duarte, A. L. F., da Costa Oliveira, G., de Castilhos, M. B. M., & Del Bianchi, V. L. (2024). Production of carotenoids by *Rhodotorula mucilaginosa* using glucose and cassava wastewater and a comparison between ethanol, methanol and DMSO in their extraction. *Biocatalysis and Agricultural Biotechnology*, 58, Article 103167. doi.org
- Dintzis, F. R., Bagley, E. B., & Felker, F. C. (1995). Shear-thickening and flow-induced structure in a system of DMSO containing waxy maize starch. *Journal of Rheology*, 39(6), 1399–1409. doi.org
- El-Masry, M. H., Khalil, A. I., Hassouna, M. S., & Ibrahim, H. A. H. (2002). In situ and in vitro suppressive effect of agricultural composts and their water extracts on some phytopathogenic fungi. *World Journal of Microbiology and Biotechnology*, 18(6), 551–558. doi.org

- Gadoury, D. M., Cadle-Davidson, L. A. N C. E., Wilcox, W. F., Dry, I. B., Seem, R. C., & Milgroom, M. G. (2012). Grapevine powdery mildew (*Erysiphe necator*): A fascinating system for the study of the biology, ecology and epidemiology of an obligate biotroph. *Molecular Plant Pathology*, 13(1), 1–16. doi.org
- Galvao, J., Davis, B., Tilley, M., Normando, E., Duchen, M. R., & Cordeiro, M. F. (2014). Unexpected low-dose toxicity of the universal solvent DMSO. *The FASEB Journal*, 28(3), 1317–1330. doi.org
- Ganchev, D. (2026). Acute algicidal toxicity of dimethyl sulfoxide towards *Cyclotella meneghiniana*. *KNOWLEDGE - International Journal*, 75(3), 277–282.
- Graeff, S., Link, J., & Claupein, W. (2006). Identification of powdery mildew (*Erysiphe graminis* sp. *tritici*) and take-all disease (*Gaeumannomyces graminis* sp. *tritici*) in wheat (*Triticum aestivum* L.) by means of leaf reflectance measurements. *Central European Journal of Biology*, 1(2), 275–288. doi.org
- Jacob, M. S. W., & Wood, P. D. D C. (1967). Dimethyl sulfoxide (DMSO) toxicology, pharmacology, and clinical experience. *The American Journal of Surgery*, 114(3), 414–426. doi.org
- Kumar, B., Gangwar, M. S., & Rathore, V. S. (1976). Effect of dimethyl sulfoxide (DMSO) on zinc availability (L-value), growth and metabolic activities of rice plants. *Plant and Soil*, 45(1), 235–246. doi.org
- Kumari, R., Ashraf, S., Bagri, G. K., Khatik, S. K., Bagri, D. K., & Bagdi, D. L. (2018). Extraction and estimation of chlorophyll content of seed treated lentil crop using DMSO and acetone. *Journal of Pharmacognosy and Phytochemistry*, 7(3), 249–250.
- Li, T., Lee, S., Choi, T., Lim, Y., Ma, T., & Lee, J. (2025). Dimethyl sulfoxide suppresses *Fusarium graminearum* pathogenicity by modulating key life cycle traits. *The Plant Pathology Journal*, 41(5), 595.
- Manjunath, P., & Shivaprakash, B. V. (2013). Pharmacology and clinical use of dimethyl sulfoxide (DMSO): A review. *International Journal of Molecular Veterinary Research*, 3(1), 1–6. doi.org
- Melhuish, J. H., Jr., & Bean, G. A. (1971). Effect of dimethyl sulfoxide on the sclerotia of *Sclerotium rolfsii*. *Canadian Journal of Microbiology*, 17(3), 429–431. doi.org
- Oliveira, S. S., Cruz, M. S. F. V., Ristau, A. C. P., Braz, H., Alves, T. N., Rocha, H. G. A., & Guimarães, V. F. (2018). Use of dimethylsulfoxide on the postharvest conservation and quality of strawberry and peach. *Journal of Experimental Agriculture International*, 26(6), 1–10. doi.org
- Osman, M. E. S., El-Shaphy, A. A. E. N., & Ayid, M. M. (2017). Evaluation of the inhibitory effect of dimethyl sulfoxide on fungal degraded archaeological wood. *International Journal of Conservation Science*, 8(3), 431–442.
- Pal, R., Mamidi, M. K., Das, A. K., & Bhonde, R. (2012). Diverse effects of dimethyl sulfoxide (DMSO) on the differentiation potential of human embryonic stem cells. *Archives of Toxicology*, 86(4), 651–661. doi.org
- Park, C. H., & Martinez, B. C. (1992). Enhanced release of rosmarinic acid from *Coleus blumei* permeabilized by dimethyl sulfoxide (DMSO) while preserving cell viability and growth. *Biotechnology and Bioengineering*, 40(4), 459–464. doi.org
- Peng, M., & Kuc, J. (1992). Peroxidase-generated hydrogen peroxide as a source of antifungal activity in vitro and on tobacco leaf disks. *Phytopathology*, 82(6), 696–699.
- Petrucelli, V., Brasili, E., Varone, L., Valletta, A., & Pasqua, G. (2020). Antifungal activity of dimethyl sulfoxide against *Botrytis cinerea* and phytotoxicity on tomato and lettuce plants. *Plant Biosystems - An International Journal Dealing with all Aspects of Plant Biology*, 154(4), 455–462. doi.org
- Piepho, H. P., Malik, W. A., Bischoff, R., El-Hasan, A., Scheer, C., Sedlmeier, J. E., & Voegelé, R. T. (2024). Efficacy assessment in crop protection: A tutorial on the use of Abbott's formula. *Journal of Plant Diseases and Protection*, 131(6), 2139–2160. <https://doi.org/10.1007/s41348-024-00968-0>
- Shinano, T., Lei, T. T., Kawamukai, T., Inoue, M. T., Koike, T., & Tadano, T. (1996). Dimethylsulfoxide method for the extraction of chlorophylls a and b from the leaves of wheat, field bean, dwarf bamboo, and oak. *Photosynthetica*, 32(3), 409–415.
- Snyder, J. C., Sowokinos, J. R., & Desborough, S. L. (1977). Rapid extraction and solubilization of potato starch with dimethylsulfoxide. *American Potato Journal*, 54(11), 513–517. doi.org
- Swanson, B. N. (1985). Medical use of dimethyl sulfoxide (DMSO). *Reviews in Clinical & Basic Pharmacology*, 5(1-2), 1–33.
- Taskeen-Un-Nisa, Bhat, M. Y., Pala, S. A., & Mir, R. A. (2011). In vitro inhibitory effect of fungicides and botanicals on mycelial growth and spore germination of *Fusarium oxysporum*. *Journal of Biopesticides*, 4(1), 53–56.
- Van Leeuwen, G. C. M., Stein, A., Holb, I., & Jeger, M. J. (2000). Yield loss in apple caused by *Monilinia fructigena* (Aderh. & Ruhl.) Honey, and spatio-temporal dynamics of disease development. *European Journal of Plant Pathology*, 106(6), 519–528. doi.org
- Wang, J., & Higgins, V. J. (2005). Nitric oxide has a regulatory effect in the germination of conidia of *Colletotrichum coccodes*. *Fungal Genetics and Biology*, 42(4), 284–292. doi.org