PRINT ISSN 1119-8362
Electronic ISSN 2659-1499

Full-text Available Online at
https://www.ajol.info/index.php/jasem
https://www.bioline.org.br/ja

J. Appl. Sci. Environ. Manage.
Vol. 28 (9) 2839-2841 September 2024

Biocontrol Potential of Endophytic Fungi against Orobanche Infestations in Fruit Crops: A Case Study in Delta State, Nigeria

***UKPENE, AO; ODUMA, EO**

Department of Biological Sciences, University of Delta, Agbor, Nigeria

**Corresponding author email: anthony.ukpene@unidel.edu.ng*
**ORCID: 0000-0002-5908-5411*
**Tel: +2348064107048*

Co-Author Email: ewoma.oduma@unidel.edu.ng

ABSTRACT: This study investigated the potential of endophytic fungi as biocontrol agents against Orobanche infestations in fruit crops such as chickpea, soybean, lentil, and pea plants from different sites in Ika agricultural habitats in Delta State, Nigeria, using appropriate standard methods. *Fusarium sp*. showed significant inhibition of *Orobanche crenata* (75%) by mycotoxin production and *Penicillium sp*. (65%) by biocombustion. Field trials showed a significant reduction in Orobanche-infested areas, with *Fusarium sp*. (40% to 10%) and *Penicillium sp*. (30% to 10%) treatments showing significant yield improvement (20% and 22%, respectively). The study concluded that selected fungi, especially *Fusarium sp.* and *Penicillium sp.*, are effective control agents for Orobanche in legume crops. This information contributes to sustainable agriculture by providing environmentally friendly alternatives to pesticides, crop yields, and economic losses from plants, and it reduces the incidence of pests.

DOI: https://dx.doi.org/10.4314/jasem.v28i9.28

Cite this Article as: UKPENE, A. O.; ODUMA, E. O. (2024). Biocontrol Potential of Endophytic Fungi against Orobanche Infestations in Fruit Crops: A Case Study from Delta State, Nigeria. *J. Appl. Sci. Environ. Manage.* 28 (9) 2839-2841

Dates: Received: Received: 07 July 2024; Revised: 15 August 2024; Accepted: 19 August 2024 Published: 05 September 2024

Keywords: biocontrol; endophytic fungi; Orobanche; parasitic plants; integrated pest management

Legume crops play an important role in global agriculture, providing essential nutrients, improving soil fertility by fixing nitrogen, and supporting food security in many developing regions, but *Orobanche spp*. (commonly known as broomrape), which are important rootworms that extract water and nutrients from their host plants, cause significant crop losses, especially in areas such as sub-Saharan Africa where agriculture is of subsistence holdings. Parasitic plants such as Orobanche have complex life cycles and complex host-parasite interactions, making them difficult to control (Hu *et al*., 2020; Mutuku *et al*., 2020). Conventional control strategies for Orobanche, such as crop rotation, deep tillage, and use of resistant varieties, have had limited success. Weed control with herbicides can be effective but often carries environmental risks and is not always economically viable for smallholder farmers, so a sustainable, environmentally friendly approach to dealing with plant pests in legume crops is urgently needed (Reckling *et al*., 2020). Biological control offers a promising alternative by using natural enemies to control plant pests. Endophytic fungi that do not harm plant roots emerged as potential biocontrol agents. These fungi can promote plant growth, improve resistance, and compete with or inhibit pest-infested plants. The quest for endophytic fungi as biocontrol agents against the legumes Orobanche offers a sustainable alternative to this agricultural challenge. Silva *et al*., 2019; Bamisile *et al*., 2021). Endobiotic

**Corresponding author email: anthony.ukpene@unidel.edu.ng*
**ORCID: 0000-0002-5908-5411*
**Tel: +2348064107048*

fungi are known to play a variety of roles in plant health. A variety of active compounds, including antibiotics, antifungals, and plant growth hormones, can be produced that can directly or indirectly affect plant pests, and even endophytic fungi can provide systemic resistance of the host plants to enhance all their defense mechanisms (Grabka *et al*., 2022; Jaber and Ownley, 2018; Dara, 2019). These component interactions make endophytic fungi suitable candidates for Integrated Pest Management (IPM) strategies aimed at controlling Orobanche in legume crops (Rubiales, 2023).

The use of plant pests in crop production has attracted considerable attention, and strategies have been developed to reduce their negative effects in agriculture. Endophytic fungi as pesticides have shown promising results in revealing plant health enhancement and plant pathogen prevention. Verma *et al*. (2022) documented that fungal endophytes enhance plant stress resilience and promote climate-resilient sustainable agriculture by boosting tolerance to various stresses and acting as biocontrol agents and biofertilizers. These fungi can inhabit one of the plant tissues to produce bioactive compounds and provide structure of energy, which can be used to control parasitic plants such as Orobanche. Recent research has focused on the identification and use of endophytic fungi to control plant pests. A study by Natsiopoulos *et al*. (2022) on *Trichoderma sp*p. lends credence and confirmation to the potential use of endophytic fungi in biocontrol of Orobanche in tomato plants. Similarly, Vurro (2023) also evaluated bioherbicide approaches for root parasitic broomrapes but opined that adviced that further scientific and technological progress for commercial success was required. All these studies suggest that endophytic fungi may act as effective biocontrol agents against the legume Orobanche, although specific methods and field efficacy remain areas of active investigation. The role of soil microorganisms in plant health and pest control has also been increasingly developed. Carrión *et al*. (2019) demonstrated that endophytic root microbiomes, which include bacteria and fungi, can suppress fungal root diseases by producing antifungal enzymes and secondary metabolites.

Despite the potential of endophytic fungi in biocontrol, there is a lack of comprehensive research focusing on their use against legume Orobanche, so most existing research focuses on the use of endophytes for infection control of soil parasites to improve plant growth under stress conditions. The specific mechanisms of endophytic fungal control of orchids and their effects on harvest health and yield remain poorly investigated (Bamisile *et al*., 2018). Addressing this knowledge gap is essential to developing useful and effective biocontrol strategies for field use. Hence, the objective of this paper was to investigate the potential of endophytic fungi as biocontrol agents against Orobanche infestations in fruit crops such as chickpea, soybean, lentil, and pea plants from different sites in Ika agricultural habitats in Delta State, Nigeria.

MATERIALS AND METHODS

Isolation and Identification of Endophytic Fungi: Legume crops (cowpea, chickpea, soybean, lentil, and pea) were collected from various locations in Ika farming communities in Delta State, Nigeria (Mbiri, Ofien, Oki, Abavo and Udomi). Root and stem samples were surface-sterilized using a series of ethanol and sodium hypochlorite washes, followed by washing with sterile distilled water and then samples were cut into small pieces and plated on potato dextrose agar (PDA) plates. The plates were incubated at 25°C for 7–10 days. Emerging fungal communities were subcultured to obtain pure isolates and then identified based on their morphological characteristics.

In vitro inhibition assay: Isolated endophytic fungi were tested for in-vitro growth inhibition of Orobanche species (*Orobanche crenata, Orobanche aegyptiaca, Orobanche ramose*, and *Orobanche minor*). Fungal culture filters and mycelium plugs were plated onto Orobanche-coated agar plates. The percentage inhibition of germination and growth in Orobanche was evaluated after 14 days. Inhibition mechanisms such as mycotoxin production, nutrient competition, and excretion of volatiles were studied by biochemical tests and microscopy.

Greenhouse Trials: Effects of selected endogenous fungi (*Fusarium sp*., *Trichoderma sp*., *Aspergillus sp*., *Penicillium sp*., and *Cladosporium sp*.) on legume growth and yield were tested under greenhouse conditions. Legume seeds were treated with fungal spore suspension prior to planting in sterilized soils. Control groups had no fungicide treatment. Plants were grown under standard conditions, and plant height, number of pods per plant, and yield data were collected. Plant health was scored visually based on leaf color, root growth, and disease symptoms.

Field Trials: Field tests were conducted in the areas from where legume samples were originally collected. Plots were established for each treatment group (control and endophytic fungus treated). Legume seeds were inoculated with fungal spores and cultivated. Orobanche invasion was monitored by the percentage of pre-treatment and post-treatment. Yield data were collected at the end of the growing season, and the percent improvement in yield was calculated.

Economic Analysis: An economic analysis was conducted to assess the cost-effectiveness of using endophytic fungi for control of Orobanche. Costs included production and application of fungal inoculation per hectare. Yields from field trials were used to calculate the increase in income due to increased yields. Net benefits were calculated by subtracting treatment expenses from the increase in income. These methods provided detailed information on the isolation, identification and efficacy of endophytic fungi on Orobanche on legumes, their effects on fruit growth and yield, and the economic benefits of such biocontrol. To perform the economic analysis of the cost-effectiveness of using endophytic fungi for the control of Orobanche, the following linear models and input-output economics equations were used, thus:

*Cost of Treatment (C)***:** The total cost of using the endophytic fungi, including both production and application costs and was computed thus:

$$C = C_{prod} + C_{app} \qquad (1)$$

Where: C_{app} = Cost of application of fungal inoculum per hectare

Increase in Income (ΔI): Field trial data were used to determine the difference in yield between treated and control plots and the increase in income computed as:

$$\Delta l = (Y_{treated} - Y_{treated}) x\, P \qquad (2)$$

Where, $Y_{control} = Yield\ per\ hecter\ with\ fungal\ treatment\ (in\ Kg$; P = market price per kilogram of the crop

Net Benefit (B): This was derived by subtracting the total cost of treatment from the income increase to get the net benefit, given as,

$$B = \Delta l - C \qquad (3)$$

Where, ΔI = Increase in income due to the treatment; C = Total cost of treatment

Benefit Cost Ratio (BCR): $(BCR) = \frac{B}{C}$ (4)

Where: B = Net benefit; C = Total cost of treatment

Return on Investment (ROI): This equation expresses the net benefit as a percentage of the treatment cost.

$$ROI = \frac{B}{C} x100\% \qquad (5)$$

$$Payback\ period = \frac{C}{\Delta L} \qquad (6)$$

This equation estimates the time required to recover the initial cost of treatment from the additional income generated.

RESULTS AND DISCUSSION

The results of the study are presented in Tables 1-5. The data presented in Table 1 highlight the isolation and identification of endophytic fungi from various legume crops within different regions used in the study. In the table, five fungal isolates, each associated with a specific leguminous plant, were distinguished. Isolate EF01 was obtained from cowpea plants collected in Mbiri farmland on May 12, 2023. The isolate was identified as *Fusarium sp*., a genus known for its diverse interactions with plants, including both pathogenic and beneficial strains. Isolate EF02 obtained from chickpea plantations in Ofien farmland was identified on June 8, 2023, as *Trichoderma sp*., a genus renowned for its biocontrol properties against numerous plant pathogens. EF03 isolates from soybean plants in Oki farmland were collected on July 14, 2023, and identified as *Aspergillus sp*., a genus that includes species with significant roles in soil health and plant growth promotion. Similarly, isolate EF04 was obtained from lentil plants in Abavo on March 21, 2023, infected with *Penicillium sp*., another genus with potential biocontrol capabilities and importance in agricultural ecosystems. EF05, isolated from pea plants in Udomi, on September 15, 2023, was finally obtained and found to be *Cladosporium sp*., which is known for its diverse ecological roles, including endophytism and potential biocontrol applications. This table provides a blueprint of endophytic fungi diversity associated with legume crops in different agroecological zones in Ika and sets the stage for further investigation of their potential as biocontrol agents against plant pathogens.

In-vitro studies on the inhibitory effects of various endophytic fungi on Orobanche species are presented in Table 2, which shows that the former can effectively be used as biocontrol agents for the latter. *Fusarium sp*., the epiphytic fungi extracted from legumes, could inhibit *Orobanche crenata* with about 75% reduction in growth due to mycotoxins produced by this fungi. This mechanism resulted in strong inhibition, suggesting *Fusarium sp*. as a potent candidate for biocontrol. *Trichoderma sp*., known for its antagonistic interactions with pathogens, exhibited a

60% inhibition against *Orobanche aegyptiaca*, indicating moderate effectiveness. Similarly, *Aspergillus sp*., through nutrient competition, achieved a 50% inhibition of *Orobanche ramose*, showing moderate inhibitory potential. *Penicillium sp*., producing volatile organic compounds, displayed a 65% inhibition against *Orobanche minor*, indicating good efficacy in suppressing parasite growth. Similarly, *Cladosporium sp*. exhibited moderate inhibition (55%) against *Orobanche crenata*, attributed to its mycoparasitic activity. It was reported by Bastías *et al*. (2021) that endophytic fungi can eliminate the plant growth-defense trade-off by stimulating plant growth hormones and producing anti-herbivore alkaloids without compromising plant growth. These findings underscore the diverse mechanisms by which endophytic fungi can impede *Orobanche* growth in vitro, paving the way for further exploration of these fungi as sustainable biocontrol agents in agricultural practices aimed at managing parasitic plant infestations in legumes.

Table 1: Endophytic fungi isolated from legume crops

Isolate Code	Legume Host Plant	Fungi Species	Location of Isolation	Date of Isolation
EF01	Cowpea	*Fusarium sp.*	Mbiri, farmlands	2023-05-12
EF02	Chickpea	*Trichoderma sp.*	Ofien farmlands	2023-06-08
EF03	Soybean	*Aspergillus sp.*	Oki farmland	2023-07-14
EF04	Lentil	*Penicillium sp.*	Abavo	2023-08-21
EF05	Pea	*Cladosporium sp.*	Udomi	2023-09-15

Table 2: In vitro inhibition of Orobanche growth by endophytic fungi

Fungi Species	Orobanche Species	Percentage Inhibition (%)	Inhibition Mechanism (Observed)	Notes
Fusarium sp.	Orobanche crenata	75	Mycotoxin production	Strong inhibition
Trichoderma sp.	Orobanche aegyptiaca	60	Antagonistic interaction	Moderate inhibition
Aspergillus sp.	Orobanche ramose	50	Competition for nutrients	Moderate inhibition
Penicillium sp.	Orobanche minor	65	Volatile organic compounds	Good inhibition
Cladosporium sp.	Orobanche crenata	55	Mycoparasitism	Moderate inhibition

Table 3: Impact of endophytic fungi on legume growth and yield

Treatment Group	Legume Host Plant	Average Plant Height (cm)	Average Pod Number	Average Yield (g/plant)	Health Status (Visual Score)
Control (No fungi)	Cowpea	30	20	50	3
Fusarium sp. Treated	Cowpea	35	25	60	4
Trichoderma sp. Treated	Chickpea	28	22	55	4
Aspergillus sp. Treated	Soybean	33	24	58	4
Penicillium sp. Treated	Lentil	29	21	52	3
Cladosporium sp. Treated	Pea	32	23	64	4

Table 3 presents the outcomes of greenhouse experiments assessing the impact of different endophytic fungi treatments on legume growth parameters and yield. The control group, untreated with fungi, exhibited an average plant height of 30 cm and produced 20 pods per plant, yielding 50 grams per plant, with a corresponding health visual score of 3. In contrast, *Fusarium sp*. showed an increase in all metrics, with an average plant height of 35 cm, 25 pods per plant, and a yield of 60 grams per plant, resulting in an improved health indicator score of 4 mean. Similarly, *Trichoderma sp.* exhibited a comparable health status score of 4, with soybean plants growing up to 28 cm, recording 22 pods per plant and a seed yield of 55 g per plant. *Aspergillus sp*. also showed remarkable growth, with an average plant height of 33 cm, 24 pods per plant, and 58 g of fruit per plant, resulting in a visual health score of 4. Factors such as plant height (29 cm) and number of seedlings (21 pods per plant) of *Penicillium spp.*, 52 grams per plant, were obtained, which gave a health status score of 3. In addition, *Cladosporium sp* showed an improved health status of 4 with an average plant height of 32 cm, 23 pods per plant, and a yield of 64 grams per plant. These results suggest that the use of endophytic fungi such as *Fusarium sp*., *Trichoderma sp*., *Aspergillus sp*., and *Penicillium sp*. can positively influence plant growth and yield under controlled greenhouse conditions. Different fungal species display different responses to host isolation, affecting their richness, abundance, and composition in root nodules as observed by Mony *et al*., (2020). Healthy soil scores reflect whole plant vigour and stress resistance, and highlight the potential of endophytic fungi as beneficial symbionts for increasing crop yields in agricultural areas.

Table 4 presents the results from field trials evaluating the efficacy of different endophytic fungi treatments in controlling Orobanche infestations in various legume crops across different locations in Ika farmlands. In Mbiri farmlands, cowpea plants treated with *Fusarium sp.* showed a notable reduction in Orobanche infestation from 40% to 10% of the area affected. This treatment corresponded with a 20% improvement in yield compared to untreated controls. Similarly, in Ofien farmlands, chickpea plants treated with *Trichoderma sp*. experienced a decrease in Orobanche infestation from 35% to 15% of the area, resulting in a 15% yield improvement. In Oki farmland, soybean plants treated with *Aspergillus sp*. demonstrated a reduction in Orobanche infestation from 45% to 20% of the area affected. This treatment led to an 18% increase in yield compared to untreated soybeans. Lentil plants in Abavo, treated with *Penicillium sp.*, exhibited a decrease in Orobanche infestation from 30% to 10% of the area, with a corresponding 22% improvement in yield. Lastly, pea plants in Udomi, treated with *Cladosporium sp.*, showed a decrease in Orobanche infestation from 25% to 10% of the area and a 16% yield improvement. These findings underscore the potential of endophytic fungi as effective biocontrol agents against Orobanche infestations in legume crops under field conditions. The significant reductions in Orobanche infestation levels observed across different locations suggest that specific fungal treatments can mitigate parasitic plant damage and enhance crop productivity, similar to the records of Poveda *et al*. (2020) that filamentous fungi like Trichoderma, mycorrhizal, and endophytic fungi can reduce plant-parasitic nematode damage and enhance crop productivity by enhancing plant defenses and providing higher nutrient and water uptake. The yield improvements associated with fungal treatments highlight their practical benefits for farmers facing Orobanche-related challenges in diverse agricultural settings.

Table 4: Field trials: Efficacy of endophytic fungi in controlling Orobanche infestation

Field Location	Legume Host Plant	Treatment (Fungi Species)	Orobanche Infestation(Pre-treatment, % area)	Orobanche Infestation (Post- treatment, % area)	Yield Improvement (%)
Mbiri, farmland	Cowpea	*Fusarium sp.*	40	10	20
Ofien farmland	Chickpea	*Trichoderma sp.*	35	15	15
Oki farmland	Soybean	*Aspergillus sp.*	45	20	18
Abavo	Lentil	*Penicillium sp.*	30	10	22
Udomi	Pea	*Cladosporium sp.*	25	10	16

Table 5 provides an economic analysis of the costs and benefits of various endophytic fungal treatments used to control Orobanche on legume crops per hectare, represented in Nigerian Naira (₦). Plot without treatment incurred no costs and showed no increased yields, resulting in lower revenue earnings and profit margins. Legumes fields treated with *Fusarium sp*. attracted costs of ₦15,000 per hectare, with an average yield of 30,000 kg per hectare. This led to an increased corresponding yield of ₦ 45,000 per hectare, thus giving a net profit of ₦ 30,000. In the same way, for *Trichoderma sp*., the treatment cost was ₦14,000/ha but the yield increased by 28,000 kg/ha. These yield improvements led to a tune of ₦ 42,000 per hectare yield and a net profit of ₦ 28,000.

Furthermore, treatment with *Aspergillus sp*. incurred the costs of ₦16000 per hectare, yielding an increase of 32000 kg/ha. Similarly, income increased by ₦48000 per hectare, with a net profit of ₦32000 after deducting treatment costs. In addition, the Penicillium *sp*. cost of treatment per hectare was ₦14500, and the average yield increased by 29000 kg/ha. This increase in yield simultaneously increased revenue by ₦43500 per hectare, resulting in a net profit of ₦29000. These findings illustrate the economic feasibility of using endophytic fungi treatments for Orobanche control in legume cultivation in Ika farmlands. The positive net benefits across all fungal treatments underscore their potential to enhance agricultural productivity and profitability, thereby promoting sustainable farming practices and resilience against parasitic plant infestations.

Table 5: Economic analysis of using endophytic fungi for Orobanche control in legumes

Treatment Group	Cost of Treatment	Average Yield Increase (kg/hectare)	Revenue Increase (₦/hectare)	Net Benefit (₦/hectare)
	Per Hectare (₦)			
Control (No fungi)	0	0	0	0
Fusarium sp. Treated	15000	30000	45000	30000
Trichoderma sp. Treated	14000	28000	42000	28000
Aspergillus sp. Treated	16000	32000	48000	32000
Penicillium sp. Treated	14500	29000	43500	29000

The study offers significant information in which endophytic fungi could be effectively applied to combat Orobanche infection in legume crops. In-vitro tests showed that Orobanche growth inhibition rates range from one fungi species to another; for example, *Fusarium sp*. demonstrated rather high inhibition through its ability of mycotoxin production, while *Trichoderma sp*. in a manner that takes place in the antagonistic interaction, *Aspergillus sp*. by competing nutrients, *Penicillium sp*. via volatile organic compounds, and *Cladosporium sp*. through mycoparasitism. These mechanisms correlate with previous studies on using fungi for managing parasitic plants, especially acknowledging that fungi use various techniques in order to combat parasitic plants. Essarioui *et al*. (2020) reported that coevolutionary antagonistic interactions between Fusarium and Streptomyces are driven by resource competition, suggesting antibiotics act as weapons in mediating bacterial-fungal interactions in soil.

It was established that there was successful isolation and characterization of endophytic fungi from the different legume crops in Ika agricultural farmlands. Some of the fungi species found in the study and their classification include; *Fusarium sp*., *Trichoderma sp*., *Aspergillus sp*., *Penicillium sp*., and *Cladosporium sp*., residing in various areas separated from others, including Mbiri, Ofien, Oki, Abavo, and Udomi. This step proved to be strategic in determining the types of fungi that are symbiotic with legumes and the opportunities for biocontrol.

Secondary, in-vitro assays proved that these fungi are effective in controlling Orobanche species. The findings revealed a highly significant decrease in the growth rate of Orobanche throughout the experimental period and the efficacy of *Fusarium sp*. and *Penicillium sp*. exhibiting particularly strong inhibition.

Greenhouse tests also corroborated the biocontrol perspective of these fungi by optimizing legume growth characteristics through treatments with Fusarium sp., Trichoderma sp., Aspergillus sp., Penicillium sp., and Cladosporium sp. The subsequent improvement in plant height, pod number, and increased plant health rating further supported the treatment's effectiveness over the controls. Thus, the obtained outcomes prove that fungal treatments can enhance legume production under controlled environmental conditions, similar to the findings of Alves *et al.* (2021), who documented that fungal treatments improve soil nutrient availability, arbuscular mycorrhizal colonization, and common bean growth.

The performance of the fungal treatments was tested on field level in different regions of the Ika agricultural landscape and revealed that the fungal treatments were effective in controlling Orobanche and increasing the yield of legumes. Decreases in Orobanche-infested area and hence an increase in yield were noted in the study after treatments with *Fusarium sp*., *Trichoderma sp*., *Aspergillus sp*., *Penicillium sp*., and *Cladosporium sp*. These outcomes evidence that it is possible and effective to combine the use of fungal biocontrol strategies for managing Orobanche pathogens and its effect on yield.

Earlier works have approximated the positive functions of the endophytic fungi in fortifying the plant health, uptake of nutrients, and containment of the plant diseases. These facts are supported by our results showing that particular endophytic fungi, such as Fusarium sp., Trichoderma sp., Aspergillus sp., Penicillium sp., and Cladosporium sp., possess prominent inhibitory effects on Orobanche species, either in the in vitro system or in field experiments. The obtained outcomes are consistent with the ideas of Aybeke (2020) and Oufensou *et al*. (2023), who focused on the biocontrol effect of fungi against Orobanche and discussed the methods of fungi's action, including mycotoxin production, competition for nutrients, and formation of volatile organic compounds.

Conclusion: This research assessed the effectiveness of endophytic fungi in controlling *Orobanche* in legume crops like cowpea, chickpea, soybean, lentil, and pea in Ika agricultural farmlands, Delta State, Nigeria. Endophytic fungi species were found to interfere with Orobanche development by reducing mycotoxin synthesis, depriving it of nutrients, releasing volatile organic compounds, or parasitizing it directly. These mechanisms were confirmed through in-vitro molecular assays, greenhouse, and field experiments, showing a decrease in Orobanche biomass and legume infection rates, along with an increase in yield. The study highlighted the fungi's potential as biofertilizers and their role in reducing chemical applications, emphasizing their importance in sustainable agriculture.

Declaration of Conflict of Interest: The authors declare no conflict of interest.

Data Availability Statement: Data are available upon request from the first author or corresponding author.

REFERENCES

Alves, G; Bertini, S; Barbosa, B; Pimentel, J; Junior, V; Mendes, G; Azevedo, L (2021). Fungal endophytes inoculation improves soil nutrient availability, arbuscular mycorrhizal colonization and common bean growth. *Rhizosphere,* 18: 100330. https://doi.org/10.1016/J.RHISPH.2021.100330.

Aybeke, M (2020). Aspergillus alliaceus infection fatally shifts Orobanche hormones and phenolic metabolism. *Braz. J. Microbiol.,* 51: 883 - 892. https://doi.org/10.1007/s42770-020-00283-4.

Bamisile, B; Dash, C; Akutse, K; Keppanan, R; Wang, L (2018). Fungal Endophytes: Beyond Herbivore Management. *Front. Microbiol.,* 9:544. https://doi.org/10.3389/fmicb.2018.00544.

Bamisile, B; Siddiqui, J; Akutse, K; Aguila, L; Xu, Y (2021). General Limitations to Endophytic Entomopathogenic Fungi Use as Plant Growth Promoters, Pests and Pathogens Biocontrol Agents. *Plants*, 10(10):2119. https://doi.org/10.3390/plants10102119.

Bastías, D; Gianoli, E; Gundel, P (2021). Fungal endophytes can eliminate the plant growth-defence trade-off. *New Phytol.*, 230(6): 2105-2113. https://doi.org/10.1111/nph.17335.

Carrión, V; Perez-Jaramillo, J; Cordovez, V; Tracanna, V; Hollander, M; Ruiz-Buck, D; Mendes, L; Ijcken, W; Gomez-Exposito, R; Elsayed, S; Mohanraju, P; Arifah, A; Oost, J; Paulson, J; Mendes, R; Wezel, G; Medema, M; Raaijmakers, J (2019). Pathogen-induced activation of disease-suppressive functions in the endophytic root microbiome. *Science*, 366: 606 - 612. https://doi.org/10.1126/science.aaw9285.

Dara, S (2019). Non-entomopathogenic roles of entomopathogenic fungi in promoting plant health and growth. *Insects*, 10(9): 277. https://doi.org/10.3390/insects10090277.

Essarioui, A; LeBlanc, N; Otto-Hanson, L; Schlatter, D; Kistler, H; Kinkel, L (2020). Inhibitory and nutrient use phenotypes among co-existing *Fusarium* and *Streptomyces* populations suggest local coevolutionary interactions in soil. *Environ. Microbiol.*, 22(3): 976-985. https://doi.org/10.1111/1462-2920.14782.

Grabka, R; d'Entremont, T; Adams, S; Walker, A; Tanney, J; Abbasi, P; Ali, S (2022). Fungal endophytes and their role in agricultural plant protection against pests and pathogens. *Plants*, 11(3): 384. https://doi.org/10.3390/plants11030384.

Hu, L; Wang, J; Yang, C; Islam, F; Bouwmeester, H; Muños, S; Zhou, W (2020). The effect of virulence and resistance mechanisms on the interactions between parasitic plants and their hosts. *Int. J. Mol. Sci.*, 21(23): 9013. https://doi.org/10.3390/ijms21239013.

Jaber, L; Ownley, B (2018). Can we use entomopathogenic fungi as endophytes for dual biological control of insect pests and plant pathogens. *Biol. Control*, 116: 36-45. https://doi.org/10.1016/J.BIOCONTROL.2017.01.018.

Mony, C., Vannier, N., Brunellière, P., Biget, M., Coudouel, S., Vandenkoornhuyse, P (2020). The influence of host-plant connectivity on fungal assemblages in the root microbiota of Brachypodium pinnatum. *Ecology*, 101, (4): e02976 . https://doi.org/10.1002/ecy.2976.

Mutuku, J; Mutuku, J; Cui, S; Yoshida, S; Shirasu, K (2020). *Orobanchaceae* parasite-host interactions. *New Phytol.*, 230(1): 46-59. https://doi.org/10.1111/nph.17083.

Natsiopoulos, D; Tziolias A; Lagogiannis I; Mantzoukas S; Eliopoulos PA (2022). Growth-promoting and protective effect of *Trichoderma atrobrunneum* and *T. simmonsii* on tomato against soil-borne fungal pathogens. *Crops*, 2(3): 202-217. https://doi.org/10.3390/crops2030015.

Oufensou, S; Hassan, Z; Balmas, V; Jaoua, S; Migheli, Q (2023). Perfume guns: potential of yeast volatile organic compounds in the biological control of mycotoxin-producing fungi. *Toxins*, 15(1): 45. https://doi.org/10.3390/toxins15010045.

Poveda, J; Abril-Urías, P; Escobar, C (2020). Biological control of plant-parasitic nematodes by filamentous fungi inducers of resistance: *Trichoderma*, mycorrhizal and endophytic fungi. *Front. Microbiol.*, 11: 992. https://doi.org/10.3389/fmicb.2020.00992.

Reckling, M; Bergkvist, G; Watson, C; Stoddard, F; Bachinger, J (2020). Re-designing organic grain legume cropping systems using systems agronomy. *Eur. J. Agron.*, 112: 125951. https://doi.org/10.1016/j.eja.2019.125951.

Rubiales, D (2023). Managing root parasitic weeds to facilitate legume reintroduction into Mediterranean rain-fed farming systems. *Soil Syst.*, 7(4): 99. https://doi.org/10.3390/soilsystems7040099.

Silva, N; Brooks, S; Lumyong, S; Hyde, K (2019). Use of endophytes as biocontrol agents. *Fungal Biol. Rev.*, 33(2): 133-148. https://doi.org/10.1016/J.FBR.2018.10.001.

Verma, A; Shameem, N; Jatav, H; Sathyanarayana, E; Parray, J; Poczai, P; Sayyed, R (2022). Fungal endophytes to combat biotic and abiotic stresses for climate-smart and sustainable agriculture. *Front. Plant Sci.*, 13: 953836. https://doi.org/10.3389/fpls.2022.953836.

Vurro, M (2023). Are root parasitic broomrapes still a good target for bioherbicide control? *Pest Manag. Sci.*, 80(1): 10-18. https://doi.org/10.1002/ps.7360.