



FUNGI TO THE RESCUE: THE ROLE OF DARK SEPTATE ENDOPHYTES (DSEs) IN PLANT ADAPTATION TO CLIMATE CHANGE

Lalitkumar L. Maurya^{*}, Khushal B. Muradi, Suvrajit Patra, Yogesh W. Wayal, Balaji K. Choudhari, Asha K. Raj

Kerala Agricultural University, Kerala, India

**Corresponding Author Mail ID: lalitkumarmaurya956@gmail.com*

Introduction

The impacts of climate change have led to prolonged and severe droughts, particularly in the Mediterranean region, where extensive farming has become nearly impossible, and intensive farming faces critical reductions in water availability (Mukherjee et al., 2018). These conditions have also disrupted plant-microorganism interactions, with drought and soil warming significantly diminishing symbiotic relationships across various ecosystems. Endophytic fungal relationships with plants are increasingly studied to better understand their interactions, the nature of their associations, and the potential benefits they provide to hosts. Dark septate endophytes (DSEs) are sterile or conidial fungal endophytes, typically isolated from healthy plants, that form melanized structures, such as inter- and intracellular hyphae and microsclerotia, in plant roots. DSEs exhibit low host specificity and are distributed across a wide geographical range. Their presence is particularly important in environments experiencing extreme abiotic stresses, such as drought, high salinity, or heavy metal contamination, where they enhance plant survival (Fig. 1) (Huertas et al., 2024). The relatively high abundance of DSEs in stressed habitats suggests that they might have an important function for host survival in these ecosystems (Likar and Regvar, 2013). Melanin, a complex polymeric compound accumulated in the cell wall of DSEs, has been widely recognized as the main responsible of the protective features of these fungi. In addition to antioxidative and thermo-protective characteristics, melanin protects hyphae from desiccation and mechanical disruption, improving cell wall rigidity,

hydrophobicity, and decreasing its permeability (Li et al., 2019). Unlike many fungi, DSEs are less affected by prolonged drought, maintaining high colonization rates even under such conditions. While DSEs may not provide as many benefits as mycorrhizal fungi, their ability to colonize plants more effectively under abiotic stress makes them crucial in such environments. However, simultaneous exposure to high temperatures and drought reduces DSE colonization rates, though it does not result in a loss of fungal diversity (Gehring et al., 2020). These unique traits highlight the critical role of DSEs in supporting plant resilience under climate change-induced stresses, making them a promising focus for further research in sustainable agriculture and ecosystem management.

A recent patent highlights the potential of *Rutstroemia calopus* as a dark septate endophyte (DSE) capable of enhancing crop growth and development under conditions of water and salinity stress. This biostimulant has demonstrated remarkable effects, including maintaining growth with an 80% reduction in fertilizer application for cucumber plants. Studies have shown that applying *Rutstroemia calopus* CG11560 with reduced fertilization led to a 33.8% increase in leaf area and a 30.43% increase in total dry weight compared to standard fertilization practices (Santos and Diánez, 2017). The benefits of DSEs like *R. calopus* are primarily linked to their biostimulatory effects, which enhance plant morphology, improve drought resistance, boost secondary metabolic activity, and increase the uptake of water, nutrients, and carbon. Additionally, DSEs elevate antioxidant enzyme activity and aid in the development of

adaptation mechanisms to cope with heavy metal stress. Their colonization triggers changes in cellular metabolism, biosynthesis, and signalling pathways, further modulating plant growth and resilience (Wu et al., 2021). These attributes make DSEs particularly valuable under abiotic stress conditions, where their presence can significantly reduce the need for irrigation and fertilizers. While their use may not always result in increased production, it offers an opportunity to minimize agricultural inputs while maintaining or enhancing yield, contributing to sustainable farming practices.

DSEs and their Connection to Drought and Salinity Mitigation

Dark septate endophytes (DSEs) play a significant role in helping plants tolerate abiotic stresses such as drought and salinity, which are increasingly prevalent due to climate change. These melanized fungi colonize plant roots and establish mutualistic relationships, where both the fungi and the host plant benefit. One of their key mechanisms in drought mitigation lies in enhancing water absorption and retention. DSEs improve root morphology by promoting the growth of fine roots, which increases the surface area available for water uptake (Bi et al., 2024). They also produce extracellular enzymes that break down organic matter in the soil, releasing bound water and nutrients, making them more accessible to the plant. Moreover, DSEs influence the plant's internal water balance by regulating osmolyte production, such as proline, which helps maintain cell turgor under water deficit conditions. In saline environments, DSEs mitigate salt stress through multiple pathways. High salt concentrations in soil often disrupt ionic balance and lead to ion toxicity, but DSEs help reduce this impact by modulating ion transport within plant roots. They enable selective uptake of essential ions like potassium while limiting the absorption of harmful ions such as sodium and chloride. Additionally, DSEs produce antioxidant enzymes, such as superoxide dismutase and catalase, which combat the reactive oxygen species (ROS) generated during salt stress (Zandi and Schnug, 2022). This reduces oxidative damage to plant cells and maintains

cellular integrity. DSEs also contribute to osmotic adjustment by enhancing the synthesis of compatible solutes, such as sugars and amino acids, which stabilize cellular functions in saline conditions.

The production of melanin in DSE hyphae further aids in stress tolerance. Melanin protects the fungal cells from desiccation and extreme environmental conditions, allowing them to remain functional in drought-prone and saline soils (DoSanto et al., 2017). This durability ensures a continuous symbiotic relationship with host plants, even under severe stress. Through these mechanisms, DSEs not only improve plant growth and survival in challenging environments but also contribute to the restoration of degraded lands and the sustainability of agricultural systems. Their potential for enhancing resilience to drought and salinity makes them a promising tool in climate-smart agriculture and ecosystem management.

Implications of DSEs in Fertilisation Reduction

DSEs hold significant potential in reducing dependence on chemical fertilizers by enhancing nutrient acquisition and utilization efficiency in plants. These fungi colonize plant roots and extend their hyphal networks into the surrounding soil, effectively increasing the root surface area for nutrient uptake. One of their primary contributions lies in mobilizing essential nutrients like nitrogen (N), phosphorus (P), and micronutrients, which are often limited in availability. DSEs produce extracellular enzymes, such as phosphatases and proteases, which break down organic matter and convert complex, unavailable forms of nutrients into simpler, plant-accessible forms (Huertas et al., 2024). For instance, they can solubilize inorganic phosphate or degrade organic nitrogen sources, reducing the need for synthetic fertilizers. In addition to mobilizing nutrients, DSEs improve nutrient-use efficiency within plants. By modulating nutrient transport pathways, they enable plants to optimize nutrient uptake and reduce wastage. This is particularly beneficial in systems where overuse of fertilizers often leads to environmental degradation, such as eutrophication and soil

acidification. Furthermore, DSEs promote the synthesis of plant growth-promoting compounds, such as phytohormones and secondary metabolites, which enhance overall plant health and productivity without the excessive input of fertilizers. Another significant benefit of DSEs is their ability to function in nutrient-poor soils, where conventional fertilizers may be ineffective or economically unsustainable. By improving the availability and cycling of nutrients within the soil ecosystem, DSEs contribute to the long-term maintenance of soil fertility. Additionally, their symbiotic relationship helps reduce the loss of nutrients through leaching or volatilization, particularly in sandy or heavily irrigated soils. The ability of DSEs to reduce the need for chemical fertilizers has far-reaching implications for sustainable agriculture. Lower fertilizer inputs can decrease production costs for farmers, reduce greenhouse gas emissions associated with fertilizer manufacturing and application, and mitigate the environmental impacts of nutrient runoff. Integrating DSEs into farming systems can thus support the transition to eco-friendly agricultural practices, ensuring food security while preserving natural resources. As a result, DSEs represent a valuable biological tool for

addressing the twin challenges of sustainable crop production and environmental conservation.

Compatibility of DSEs with Other Microorganisms

DSEs exhibit strong compatibility with other microorganisms, forming microbial networks that benefit plant and soil health. They often coexist with beneficial microbes like mycorrhizal fungi, nitrogen-fixing bacteria, and plant growth-promoting rhizobacteria (PGPR), engaging in synergistic interactions rather than competition. For instance, DSEs complement arbuscular mycorrhizal fungi (AMF) by thriving in harsher conditions where AMF may be less effective, collectively improving nutrient and water uptake and enhancing stress tolerance (Santos *et al.*, 2021). DSEs also work alongside nitrogen-fixing bacteria like *Rhizobium* and *Azospirillum* to optimize nitrogen availability, with DSEs mobilizing nutrients and bacteria supplying fixed nitrogen, reducing the need for synthetic fertilizers. Additionally, DSEs interact with PGPR, such as *Pseudomonas* and *Bacillus*, which promote plant growth and suppress pathogens. These collaborations create a multi-functional microbial consortium that boosts plant

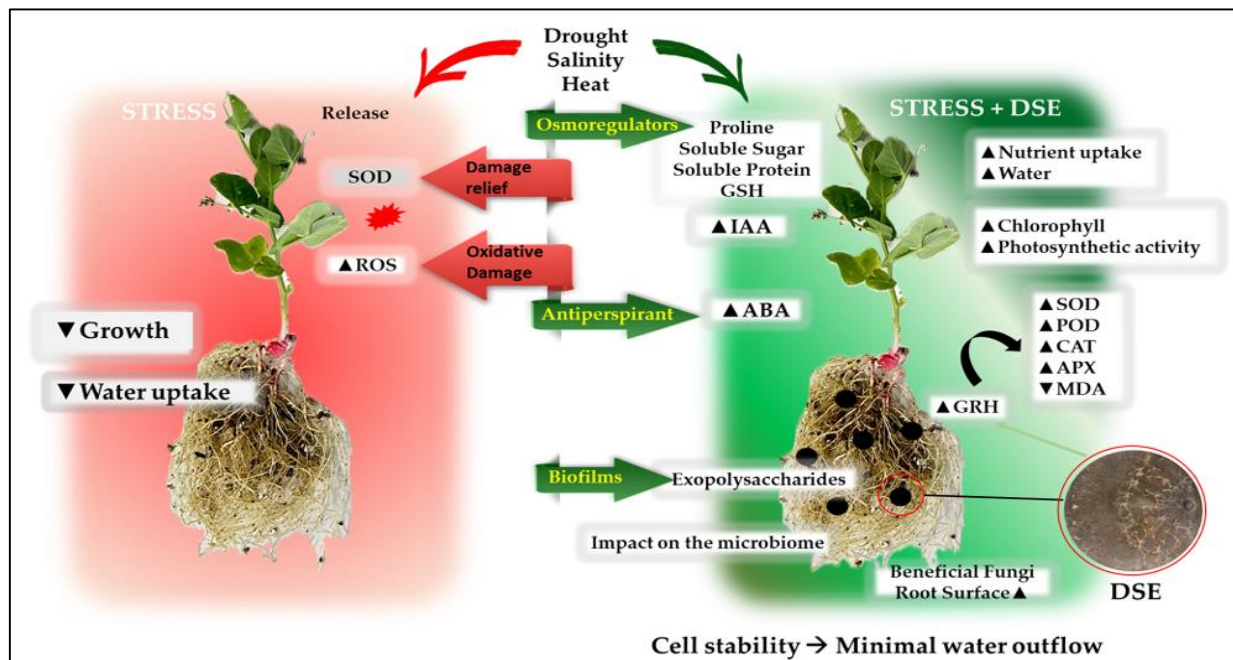


Figure 1. Schematic representation regarding the different effects and mechanisms of action reported with DSE inoculation in plants (retrieved from Huertas *et al.*, 2024).

productivity and resistance to stresses. By producing extracellular enzymes and secondary metabolites, DSEs support microbial diversity and soil health. Melanin production enhances soil organic matter, benefiting other soil microbes, while biofilm formation with other microorganisms improves microbial survival and nutrient cycling (Huertas et al., 2024). This compatibility highlights DSEs' potential in integrated microbial inoculants for sustainable agriculture, fostering resilient ecosystems and supporting modern agricultural practices through multi-functional biofertilizers and biostimulants.

Compatibility of DSEs with Active Chemical Substances

DSEs demonstrate resilience and adaptability in chemically stressed environments, making them compatible with agricultural practices involving fertilizers, pesticides, and herbicides. Their robust traits, including melanized hyphae, antioxidant enzymes, and the ability to metabolize complex compounds, enable them to thrive despite chemical inputs. DSEs contribute to soil detoxification by producing enzymes like peroxidases and laccases, breaking down toxic compounds such as pesticides and heavy metals, thereby maintaining a healthier rhizosphere (Santos et al., 2021). DSEs also enhance nutrient uptake efficiency, reducing the need for excessive fertilizer use and mitigating issues like leaching and eutrophication (Huertas et al., 2024). Their tolerance to reactive oxygen species generated by chemical inputs ensures continued functionality and plant association. Additionally, DSEs coexist with certain biocides, using their melanized structures for protection against antimicrobial agents.

However, their compatibility depends on the type and concentration of chemicals, as excessive exposure may suppress activity. Integrating DSEs into agriculture requires judicious chemical use, offering potential for sustainable practices by combining biological inoculants with minimal agrochemical inputs. This approach can improve plant productivity, reduce environmental impact, and support long-term soil health.

Conclusion and Future Directions

DSEs play a pivotal role in mitigating abiotic stresses such as drought, salinity, and nutrient deficiencies, enhancing plant resilience and productivity in challenging environments. Their ability to improve nutrient uptake, water absorption, and oxidative stress management makes them valuable allies in addressing the adverse impacts of climate change. Integrating DSEs into climate-smart agricultural practices offers a sustainable solution for improving crop yields, reducing reliance on chemical fertilizers, and restoring degraded soils. Future research should focus on unravelling the ecological interactions of DSEs with diverse plant species, understanding their genetic mechanisms of stress tolerance, and optimizing their practical applications in large-scale farming. Expanding knowledge in these areas will enable the development of tailored DSE-based bioinoculants, fostering sustainable agriculture and contributing to global climate change adaptation strategies.

References

- Huertas, V., Jiménez, A., Diáñez, F., & Santos, M. (2024). Importance of Dark Septate Endophytes in Agriculture in the Face of Climate Change. *Journal of Fungi*, 10(5), 329.
- Santos, M.; Diáñez, F. (2017). Strain of *Rutstroemia calopus*, Compositions and Uses. Patent ES2907599.,
- Do Santos, S.G.; Silva, P.R.A.; Garcia, A.C.; Zilli, J.É.; Berbara, R.L.L. (2017). Dark septate endophyte decreases stress on rice plants. *Braz. J. Microbiol.*, 48, 333–341.
- Mukherjee, S.; Mishra, A.; Trenberth, K.E. (2018). Climate change and drought: A perspective on drought indices. *Curr. Clim. Chang. Rep.* 4, 2.
- Gehring, C.; Sevanto, S.; Patterson, A.; Ulrich, D.E.M.; Kuske, C.R. (2020). Ectomycorrhizal and dark septate fungal associations of pinyon pine are differentially affected by experimental drought and warming. *Front. Plant Sci.* 11, 582574.
- Wu, F.L.; Li, Y.; Tian, W. (2021). A novel dark septate fungal endophyte positively affected

blueberry growth and changed the expression of plant genes involved in phytohormone and flavonoid biosynthesis. *Tree Physiol.* *40*, 1080–1094.

- Santos, M.; Cesanelli, I.; Diáñez, F.; Moreno-Gavira, A. (2021). Advances in the role of dark septate endophytes in the plant resistance to abiotic and biotic stresses. *J. Fungi*, *7*, 939.
- Bi, Y.; Wang, S.; Song, Y.; Christie, P. (2024). Effects of a dark septate endophyte and extracellular metabolites on alfalfa root exudates: A non-targeted metabolomics análisis. *Physiol. Plant.*, *176*, e14165.
- Zandi, P.; Schnug, E. (2022). Reactive oxygen species, antioxidant responses and implications from a microbial modulation perspective. *Biology*, *11*, 155.
- Li, X., He, C., He, X., Su, F., Hou, L., Ren, Y., & Hou, Y. (2019). Dark septate endophytes improve the growth of host and non-host plants under drought stress through altered root development. *Plant and Soil*, *439*, 259-272.