SUMMARY OF PhD THESIS

Effect of the vesicular-arbuscular fungus *Rhizophagus irregularis* on the ornamental value and bioactive compounds of the purple coneflower (*Echinacea purpurea*)

PhD student **Martin Iakab**

Scientific coordinator **Prof. Francisc Vasile Dulf, PhD**

SUMMARY

Medicinal plants represent an important group of cultivated species due to their content of biologically active compounds and their widespread use in traditional, and modern healthcare. Among these plants, *Echinacea purpurea* (L.) Moench occupies a prominent position as one of the most widely used medicinal species, mainly associated with immune-support products. In addition to its medicinal importance, *E. purpurea* is also valued as an ornamental plant, where plant height, branching and flowering intensity determine its visual appearance and horticultural value. Both medicinal quality and ornamental value are strongly influenced by cultivation conditions, including soil type, cultivation system and environmental factors.

Arbuscular mycorrhizal fungi form symbiotic associations with the roots of most terrestrial plants and have been widely studied in relation to nutrient uptake, plant growth and secondary metabolism. In medicinal plants, mycorrhizal inoculation has been reported to influence the accumulation of bioactive compounds; however, the reported effects differ between studies. A major limitation of existing research is that many experiments are conducted under highly controlled conditions and focus on short-term plant responses, which may not adequately reflect plant–fungus interactions under practical cultivation conditions. In natural or agricultural soils, introduced mycorrhizal fungi must interact with native microbial communities, and plant responses may change over time as symbiosis develops.

In this context, the present Ph.D. thesis aimed to investigate the effects of arbuscular mycorrhizal inoculation on *E. purpurea* under cultivation conditions that are closer to horticultural and agricultural practice, within the framework of sustainable plant biotechnologies and modern cultivation technologies. The research focuses on *Rhizophagus irregularis*, one of the most frequently applied arbuscular mycorrhizal fungal species in experimental studies, due to its wide host range and well-documented colonization behaviour. By integrating short-term and long-term observations and comparing greenhouse and open-field cultivation, the thesis addresses limitations of simplified experimental approaches and provides information relevant to practical cultivation systems, supported by modern biotechnological approaches. This integrated experimental approach made it possible to examine plant responses at different levels, combining aboveground and belowground observations with biochemical measurements. By applying the same parameters under different cultivation systems and over an extended period, the study provided a consistent basis for assessing the effects of mycorrhizal inoculation under conditions relevant to medicinal and ornamental plant cultivation.

The main objectives of this PhD thesis were:

The central hypothesis of this Ph.D. thesis is that inoculation with *Rhizophagus irregularis* can influence plant growth, ornamental value and bioactive compound accumulation of *Echinacea purpurea* under cultivation conditions representative of practice, and that these effects depend on soil type, cultivation system and experimental duration. Based on this hypothesis, specific

objectives were defined to guide the experimental work and to allow a structured evaluation of mycorrhizal effects under field-relevant cultivation systems. These objectives are:

O1. To evaluate the effects of *Rhizophagus irregularis* inoculation on the growth and development of *Echinacea purpurea* under both greenhouse (plants grown in pots filled with either autoclaved peat or natural soils with established soil microbiota using different soil types) and open-field conditions.

O2. To assess the influence of mycorrhizal inoculation on morphological parameters related to ornamental value, including plant height, branching, number of inflorescences and biomass production.

O3. To investigate the effects of mycorrhizal inoculation on bioactive parameters, including phenolic acid content, essential oil yield and composition, photosynthetic pigment content and enzyme activity.

O4. To compare plant responses to mycorrhizal inoculation under greenhouse pot experiments and open-field cultivation systems.

O5. To evaluate short-term and long-term effects of mycorrhizal inoculation by monitoring plant responses over two consecutive growing seasons.

The achievement of these objectives allows a comprehensive evaluation of plant–fungus interactions under cultivation systems relevant to medicinal and ornamental plant production and provides a basis for interpreting both immediate and delayed mycorrhizal effects under different environmental conditions and contributes to the development of biotechnology-oriented and modern technology-supported cultivation strategies.

The general conclusions of this PhD thesis were:

1. The results demonstrate that the effects of *Rhizophagus irregularis* inoculation on *Echinacea purpurea* were strongly influenced by cultivation conditions, soil type and experimental duration.
2. Mycorrhizal inoculation did not enhance biomass production or morphological parameters during the early stages of cultivation; however, delayed positive effects were observed on bioactive compound accumulation during the second growing season, without negatively affecting biomass production.
3. The results indicated that *Rhizophagus irregularis* influenced the qualitative composition of essential oils in *Echinacea purpurea*, with effects depending on cultivation conditions. Changes in essential oil profiles were observed in both aerial parts and roots, and were related to differences between pot and open-field cultivation as well as to soil type.
4. Root analyses demonstrated that *Rhizophagus irregularis* was able to establish and maintain mycorrhizal colonization in biologically complex soils; however, the degree of colonization varied depending on soil type and cultivation system.
5. From a practical perspective, the findings support the potential application of mycorrhizal inoculation in the cultivation of *Echinacea purpurea*, particularly in biotechnology-oriented and modern technology–supported production systems, when the primary objective is to improve bioactive compound content rather than biomass production.

Further studies should focus on the evaluation of different arbuscular mycorrhizal fungal species and inoculation strategies under field conditions and over longer cultivation periods. Additional research integrating modern biotechnological approaches may contribute to a better understanding of plant–fungus interactions and support the optimization of cultivation strategies for medicinal and ornamental plants under practical agricultural conditions.

Originality and personal contributions:

The originality of this Ph.D. thesis lies in the applied experimental design and in the integrated evaluation of arbuscular mycorrhizal effects on *Echinacea purpurea* under cultivation conditions relevant to practice. The research combines greenhouse and open-field experiments within a multi-year framework, allowing the assessment of both short-term and long-term mycorrhizal effects, which are rarely addressed in studies based on short-term or highly controlled experimental systems.

A major original contribution of the thesis is the evaluation of mycorrhizal inoculation in biologically complex soils, where *Rhizophagus irregularis* interacts with native soil microbial communities. This approach provides information on the establishment, persistence and competitive ability of the introduced fungus under conditions representative of practical horticultural and agricultural cultivation.

The thesis further contributes original results through a complex, multi-parameter assessment, integrating plant growth, ornamental characteristics and bioactive compound-related parameters within a single experimental framework. By simultaneously evaluating morphological traits, biomass production, phenolic derivatives and volatile oil composition, the study offers a comprehensive view of plant responses that goes beyond biomass-centred approaches commonly applied in mycorrhizal research.

From an applied perspective, the research provides original contributions by demonstrating that mycorrhizal inoculation can improve the biochemical quality of *Echinacea purpurea* without negatively affecting plant growth, supporting its use in biotechnology-oriented and modern technology–supported cultivation systems.