

4.1 = INTERACTION BETWEEN ENDOPHYTIC BACTERIAL COMMUNITIES ISOLATED FROM THE STEM/LEAF AND ROOT COMPARTMENTS OF THE MEDICINAL PLANTS *ECHINACEA PURPUREA* AND *ECHINACEA ANGUSTIFOLIA*.

VALENTINA MAGGINI^{1,2,3}, EUGENIA ROSARIA GALLO^{1,2}, ELISANGELA MICELI³, SAURO BIFFI⁴, ALESSIO MENGONI³, RENATO FANI³, FABIO FIRENZUOLI¹ AND PATRIZIA BOGANI³

¹Department of Experimental and Clinical Medicine, University of Florence, Largo Brambilla, 3, 50134 Florence, Italy; ²Center for Integrative Medicine, Referring Center for Phytotherapy, Tuscany Region, Careggi University Hospital, School of Medicine of Florence, 50139 Florence, Italy; ³Department of Biology, University of Florence, Via Madonna del Piano 6, 50019 Sesto Fiorentino, Italy; ⁴Botanical Garden, Casola Valsenio, Ravenna, Italy

Echinacea has arisen as a herbal medicine in the treatment of common cold and upper respiratory infections, reducing duration and/or severity of symptoms [1]. Two species, *Echinacea purpurea* (L.) Moench and *Echinacea angustifolia* (DC.) Hell, are widely used; they are rich in various phytochemicals including phenolic compounds, such as phenylpropanoids or caffeic acid derivatives (CADs), flavonoids, terpenoids, lipids, nitrogen-containing compounds and polysaccharides. The concentrations of these bioactive compounds are species-specific and they vary due to geographical location, stage of plant development, time of harvest, growth conditions, processing and extraction methods. Therefore, plant cell and organ cultures have been transformed into appealing options for the production of biomass and phytochemicals. To fix the variability of crucial active substances in *Echinacea* spp, different *in vitro* methods have been developed. Recently, several studies have focused on the presence of bacterial endophytic communities in many species of aromatic and medicinal plants [2-4] to shed some light on the role that endophytes might have in the production of (plant) bioactive molecules. A previous study conducted in the Department of Biology allowed to achieve a collection of more than 500 cultivable bacterial isolates from the stem/leaf and root compartments of *Echinacea purpurea* and *E. angustifolia*, grown in the same soil and collected in the botanical garden of Casola Valsenio (Italy) [2]. The molecular analyses have showed that different communities inhabited the two plant species and different compartments of the same plant, and the low degree of strain sharing suggested the existence of a strong selective pressure within plant tissues.

The present work aimed to understand if these distinct bacterial communities could account for the differences in the medicinal properties of the two plants. To this purpose, cultures of axenic plants (in the absence of endophytes) derived from sterilized seeds were set up. After two months from germination, *E. purpurea* and *E. angustifolia* plants were infected with different bacterial endophytic strains, isolated from the stem/leaf and/or roots of *E. purpurea*. For each species, a suitable group of control plants (not infected) was provided. Plants were examined at different times for the presence of bacteria at the level both of the roots and the aerial part. Furthermore, in order to determine the possible role of bacteria as plant growth promoting, growth and physiology of plants were evaluated analyzing different parameters such as plant height and fresh weight, length of the roots and number of leaves. The same experiments were carried out on *Nicotiana tabacum* L. cv 'Xanthi' chosen as non-host species.

The overall analysis of the preliminary results suggested that the endophytic strains tended to recolonize the native niche (*i.e.* strains isolated from the leaf came back in the leaf compartment of axenic plants of the same species after *in vitro* infection). Several endophytes had a beneficial effect on the growth of the plant due to a significant increase in the number of leaves. Non-host plants did not show similar effects. In particular, the bacterial strains colonized mainly the roots of *E. angustifolia* infected with *E. purpurea* stem/leaf endophytes suggesting that the colonization of endophytic bacteria might depend on specific physiological characteristics of the host plant. On the other hand, the analysis of the *in vitro* morphogenetic behaviour indicated that the two *Echinacea* species had a different content of endogenous plant growth regulators. *E. purpurea* was able to regenerate new shoots in culture media enriched with high content of cytokinins while *E. angustifolia* produced only clusters of undifferentiated cells (callus). Then, both the presence of different endophytic communities and a different composition in the secondary metabolites (strictly related to the therapeutic properties) in the two species could depend on differences in plant primary metabolism. In this regard, experiments to characterize the chemical profile of the control and infected plants will be set up.

1) H.N. Murthy, Y.S. Kim, Park, K.Y. Paek. *Appl Microbiol Biotechnol* (2014); 98(18):7707-17

2) C. Chiellini, I. Maida, G. Emiliani, A. Mengoni, S. Mocali, et al. *Int Microbiol* (2014); 17(3):165-74.

3) S. Compant, B. Mitter, J.C. Colli-Mull, H. Gangl, A. Sessitsch. *Microbiol Ecol* (2011); 62:188-197.

4) A.C. Ikeda, L.L. Bassani, D. Adamoski, D. Stringari, V.K. Cordeiro et al. *Microb Ecol* (2013); 65:154-160.