## Stage I/2022. Polyphenols extraction and their encapsulation into mesoporous matrices for enhanced stability

## Abstract

In the stage of the project, all foreseen activities were carried out, all the objectives with promising results obtaining and all indicators were fulfilled. Hence, 10 polyphenolic ethanolic and hydroethanolic extracts from bilberry leaves and 6 from bilberry fruits were obtained by conventional and ultrasound-assisted extraction and a new extraction method was proposed under argon pressure. The influence of extraction conditions was studied both in the case leaves and fruits.

The polyphenolic extracts obtained were analysed by spectrophotometric analyses and HPLC-PDA. Polyphenolic extracts from bilberry fruits were found to be rich in flavonoids, and the leaf extracts were rich in chlorogenic acid and rutin hydrate. Extraction under optimized conditions led to obtaining extracts with high antioxidant activity.

The antibacterial activity of selected bilberry leaf extracts was evaluated on *Staphylococcus aureus* (ATCC 25923) and *Bacillus cereus* (ATCC 11778) Gram-positive strains, and on *Pseudomonas aeruginosa* (ATCC 27853) Gram-negative strain and compared with that of the main components, chlorogenic acid and rutin hydrate. A synergistic effect has been observed between chlorogenic acid and rutin hydrate present in extracts. Inhibition of microbial growth was considerably higher for the bilberry extracts compared to the standard compounds present in the same amounts as in the extract.

Ethanol and acidified ethanol were chosen for the preparation of bilberry fruit extracts. Polyphenolic bilberry extract prepared in acidified ethanol under gas pressure extraction had the highest flavonoid content.

Selected bilberry extracts were encapsulated in mesoporous silica pristine and functionalized with organic, carboxyl, mercaptopropyl groups, as well as with thiol groups and coated with fucoidan, a polysaccharide with immunotherapeutic and anticancer effects. The stability of the extracts free and encapsulated in MCM-41 silica was studied.

Biological evaluation of bilberry leaf extracts free and encapsulated in two mesoporous silica supports functionalized with carboxyl groups or proline moieties showed good cytocompatibility on the keratinocyte HaCat cell line and an antitumoral effect on skin carcinoma Mel-Juso cells. Also, the obtained data indicated the anti-inflammatory potential of both free and encapsulated bilberry leaf extract with reduced levels of IL-1 $\beta$  and TNF- $\alpha$  compared to the positive control (treated with lipopolysaccharide). The strongest anti-inflammatory activity was recorded for the treatment with the extract encapsulated in MCM-COOH.

The results obtained in this project stage were disseminated in 4 international conferences and 2 ISI papers (one only submitted).