

Streszczenie rozprawy doktorskiej w języku angielskim

Phenolic compounds are one of the most numerous groups of plant secondary metabolites that are synthesized by plants under environmental stress. Due to their chemical structure phenolic compounds are characterized by a wide spectrum of biologically active properties. One of the most important in the context of the problems of modern medicine is antibacterial activity. Due to the increasing drug resistance of bacteria, new sources of chemical compounds are sought, including secondary metabolites contained in plant tissues, which have antibacterial potential and do not induce resistance among the target microorganisms.

Venus flytrap (*Dionaea muscipula* J. Ellis) is a carnivorous plant which is the monotypic genus within the Droseraceae family. Due to its specific adaptation to the environment, the Venus flytrap has the ability to synthesize and accumulate large amounts of various phenolic derivatives: phenylpropanoids, flavonoids, anthocyanins or 1,4-naphthoquinones. For this reason, it is a useful model for research into the synthesis of phenolic compounds under stress and their biologically active properties.

The presented doctoral dissertation consists of a series of three thematically related original scientific publications. The main topic of the published works was the synthesis of phenolic compounds in tissue cultures of Venus flytrap plants, which were subjected to elicitation and genetic transformation. At the same time, the subject of the research was to study the physiological response of *D. muscipula* plants to genetic transformation and to determine the properties of biologically active secondary metabolites contained in extracts obtained from plants in the process of elicitation and transformation.

The main objectives of the dissertation were: (1) to develop an effective method of eliciting phenolic compounds in the tissue cultures of the Venus flytrap in order to increase the antioxidant and antibacterial activity of extracts obtained from the studied plants, (2) to obtain transformed *D. muscipula* plants through the use of vector transformation – *Rhizobium rhizogenes*, (3) evaluation of transformed Venus flytrap clones in terms of the accumulation of phenolic compounds and their antibacterial properties, (4) examination of selected physiological parameters of Venus flytrap plant responses to genetic transformation.

To achieve the research objectives, a biotic elicitor was used in the form of a lysate from *Cronobacter sakazakii* bacteria, at a concentration of: 1.5, 2.5 and 5% in *D. muscipula* liquid culture. The genetic transformation was performed using three strains of *R. rhizogenes* LBA

9402, ATCC 15834 and A4, which have the natural ability to incorporate a plasmid DNA fragment (T-DNA) into the plant genome. Plants of the Venus flytrap, subjected to elicitation and transformation, were studied in the context of the accumulation of biomass, phenolic compounds (using spectrophotometric methods and high-performance liquid chromatography) and their antioxidant and antibacterial properties in relation to antibiotic-resistant bacteria: *Staphylococcus aureus*, *Enterococcus faecalis*, *Escherichia coli* and *Pseudomonas aeruginosa*. The genetic transformation of *D. muscipula* was confirmed on the molecular level by means of the PCR reaction. The presence of the *rolB* gene was demonstrated in the genome of the obtained teratomas (transformed shoots) of the Venus flytrap. In addition, it was shown using Southern blot hybridization that the bacterial *rolB* gene was integrated into the DNA of the Venus flytrap clones in a single copy. The transformed clones were examined for their changes in the accumulation of malonaldehyde (an oxidative stress marker), proline, carotenoids, phenolic acids, glutathione, and antioxidant enzyme activity compared to untransformed plants. Moreover, the content of lipids and sugars, which are a component of primary metabolism, was determined in teratomas.

The obtained results showed that: (1) the efficiency of elicitation of phenolic compounds in Venus flytrap tissue cultures with the use of *C. sakazakii* lysate depends on the concentration and duration of the elicitor, (2) the applied elicitation strategy effectively increases the antioxidant and antibacterial properties of *D. muscipula* plants, (3) the bacterial *rolB* gene in the genomic DNA of Venus flytrap plants may act as an endogenous elicitor of phenolic compounds and thus lead to an increase in the antibacterial properties of extracts obtained from transformed organisms, (4) the transformation of *D. muscipula* plants causes a pleiotropic effect, leading to changes in the reactive oxygen, primary and secondary metabolism of plants, (5) genetically stable transformed organisms can be a good model for research on the synthesis and accumulation of phenolic compounds in medicinal plants.