Summary

Production of proteins of biomedical importance, including plant-derived antigens, is a promising alternative to bacterial and yeast expression systems. However, the use of transgenic plants is often associated with the problem of insufficient expression and legislation issues. In addition, the use of plant-derived oral vaccines encounters a barrier of inadequate immune response, including oral tolerance. The proposed and currently tested solution to mentioned problems is the use of purified proteins as injection vaccines and freeze-dried plant material as an orally delivered form.

As a result of the previous research, herbicide-resistant lines of transgenic lettuce plants stably producing individual HBV proteins, including the HBcAg core antigen (Hepatitis B core Antigen), were obtained. In this study, previously obtained transgenic plants and plants after agroinfiltration - one of the methods of transient expression optimized for this particular protein, were used.

Transient expression technology is a promising method of production of large amounts of biopharmaceuticals with relatively low costs. The vectors used for transient expression allow effective replication and transcription of the introduced gene in host cells and the high level of protein expression. Vectors, primarily based on the pEAQ plasmid kindly provided by prof. G. P. Lomonossoff (John Innes Center, Great Britain) were used in this research. These vectors allow the efficient expression of HBV antigens - especially the HBcAg, followed by assembly of chimeric particles with attached epitopes of the HBV surface antigen and other proteins in the host plant *Nicotiana benthamiana*.

Antigens derived from transgenic plants and after transient expression were purified using optimized procedure. Several methods of physical purification of particles from a plant extract have been tested. As a result of these studies, purification method of particles for microscopic analyzes, including electron microscopy, has been established. The final stage of the project is verification of the immunogenicity of the obtained preparations in the animal model. The results of the experiments in mice indicate a high immunogenicity of the obtained particles, comparable to commercial preparations and their ability to activate cellular and humoral immune responses.

In addition, purified capsid-like particles derived via transient expression were used as a component of bionanoparticles in the study carried out jointly with the team of prof. Michael Giersig in Centre for Advanced Technologies AMU. The particle-assembled HBcAg exhibits the ability to coat functionalized iron oxide nanoparticles, and the possibility of modifying the surface of the particle indicates their potential use for the organo-specific targeting of nanoparticles, among others for imaging or cancer therapies.