## **Abstract**

The habilitation thesis entitled "FREE AND IMMOBILIZED BIOCATALYSTS - BIOTECHNOLOGICAL APPLICATIONS" brings together the main results of scientific and academic research in the field of enzymatic biocatalysis, from the defense of the doctoral thesis to the present.

The paper consists of two main parts, A. Scientific, professional and academic achievements and B. Development plan for teaching, scientific and research careers.

The research results are structured on two major chapters, Immobilized Biocatalysts and Enzymatic bioconversion of natural substrates into products of industrial interest.

The scientific results presented in the chapter Immobilized Biocatalysts have as starting point the doctoral thesis, and based on the experience gained in protease immobilization, I extended my research to other hydrolytic enzymes, such as amylases and microbial cellulases. I chose to study these enzymes because they are very important enzymes with industrial applications.

The biotechnology industry requires eco-friendly and stable biocatalysts, operational in different conditions, easily separable and reusable. Based on this, the immobilization research lead by me aimed to find simple and efficient methods, with low cost, to stabilize microbial enzymatic preparations with protease, amylase and cellulase activity, with high immobilization yields. The main objective was to obtain insoluble enzyme preparations with high enzymatic activity, capable of catalyzing the hydrolysis reactions of their natural macromolecular substrates, with potential applications as additives in animal feed, and not only.

To increase the catalytic efficiency of immobilized enzyme preparations, I tried to combine different immobilization methods, to benefit from the advantages presented by them. I used the simplicity of the immobilization method by physical binding and the high immobilization yields that it offers in synergy with the protection ensured by the porous silica matrices obtained by the sol-gel technique. I also combined two gels, an organic-alginate gel, with an inorganic-silica gel, to obtain an immobilized preparation with higher enzymatic activity and mechanical strength.

The physico-chemical properties of biomaterials used to immobilize enzymes with macromolecular substrates influence the enzymatic activity of immobilized preparations. For positive influence, we looked for favorable variants for the synthesis of sol-gel matrices. An important role in matrices properties is played by the precursors used for the synthesis of silica gels and alcohols used as co-solvent in soil formation. Hydrophilic matrices have proven to be more suitable for obtaining immobilizers with as high enzymatic activity as possible, and the ethylene glycol is a more protective co-solvent for the enzyme than ethanol. The pore-forming agents (such as glucose or polyvinyl alcohol), the temperature at which gelation is performed, or the time of completion of the formation of the three-dimensional network are factors that influence the behavior of immobilized enzymes.

Immobilized enzyme preparations obtained by optimized methods have been shown to be stable in an environment similar to that of mammalian stomachs, which makes them usable as feed additives to improve food availability and of nutrient intake. Sol-gel biocomposites with cellulase activity have been shown to be effective as enzyme reservoirs from where the enzymes can be gradually released.

My research in this field has made it possible to win two research projects in competitions launched by the Ministry of Education and Research, and then the publication of 16 indexed papers Web of

Science, 24 papers in indexed journals BDI or CNCSIS, 8 papers were presented in conferences, and I have published 7 books and monographs.

The second chapter entitled "The enzymatic bioconversion of some natural substrates in products of industrial interest,, includes the results of the researches in the field of enzymatic hydrolysis of some natural substrates from vegetal materials.

Enzymatic hydrolysis of proteins from natural plant matrices was done to obtain a biofertilizer enriched in peptides and amino acids. Enzymatic hydrolysis of vegetable proteins has great potential given the generous supply and diversity of plants used as a protein source and compatibility with organic farming.

As a natural source of amino acids for organic farming I used less conventional matrices and plant and microbial enzymes. Plant materials and by-products are valuable sources of protein and amino acids. The transformation and recycling of by-products resulted from agriculture, through green technology, into valuable products with biofertilizing activity is achieved.

The development of this research direction was carried out within a research project funded by the Ministry of Education and Research. Based on the research results, I applied for a patent at Romanian State Office for Inventions and Trademarks. I presented the research results in 5 invention and innovation international events organized in our country and abroad.

In the last part of the thesis the plan of the evolution and development of my professional and academic career, based on my previous experience, is presented. The future projects will continue the already started research, but I will also address other new directions in the field of enzymatic biocatalysis, interconnecting the educational activities with the laboratory research and technology transfer.