

Succinic acid is one of the major chemical compounds involved in the tricarboxylic acid cycle (TCA), also known as the Krebs cycle. This cycle is a stage of metabolic processes that take place in the cells of organisms in aerobic conditions. In turn, succinic acid in anaerobic organisms can also be the end product in three biochemical pathways, *i.e.*, the reductive branch of the Krebs cycle, glyoxylate cycle, and Krebs cycle with inactive succinate dehydrogenase enzyme. The structure of the succinic acid molecule makes it one of the 12 most desirable industrial chemicals. In addition, this metabolite exhibits a variety of pro-health properties and is widely applied in the pharmaceutical industry, including the production of vitamins, amino acids and anticancer drugs.

Currently, industrial production of succinic acid is based on chemical synthesis. However, in view of attempts to manage waste biomass and eliminate environmental contaminations emitted by industrial plants, biotechnological production of succinic acid, using efficient microbial producers, has become a global point of interest. The number of microorganisms that have the ability to overproduce this metabolite is still small. Moreover, most of the identified strains are already restricted by the ability to metabolize only simple carbon sources, such as glucose. Meantime, it is important to use microorganisms characterized by the efficient production of succinic acid on complex carbon sources, such as lactose and glycerol, which are often present in waste biomass.

The purpose of the proposed research is to characterize the genetic, biochemical and physiological characteristics of the new LU1 strain of *Enterobacter* sp. characterized by the ability to produce succinic acid in the presence of lactose and glycerol. Microorganisms showing the capacity to overproduce succinic acid, *i.e.*, *Basfia succiniproducens*, *Mannheimia succiniproducens*, *Actinobacillus succinogenes*, *Anaerobiospirillum succiniciproducens* are well physiologically and biochemically characterized. Moreover, genomic sequences of these microorganisms are already known, allowing advanced research using metabolic engineering techniques. In turn, the strain identified by us was the only known bacterial strain of the genus *Enterobacter* capable of efficient succinic acid production, and hence subject to RP patent protection. Therefore, it is extremely important to conduct research that will explain regulation mechanisms of fermentation product biosynthesis, with particular emphasis on succinic acid.

Under this project, it is planned to sequence the genome of the strain *Enterobacter* sp. LU1 and conduct thorough genetic characterization of this microorganism. The resulting genomic sequence will be published in the NCBI GenBank database, being an excellent material for other researchers for further basic research on the *Enterobacter* sp. LU1 strain and phylogenetic analysis. Furthermore, the proteomic and biochemical analysis of the test strain will be performed as well as the expression analysis of selected genes involved in succinic acid biosynthesis under the influence of different carbon sources in the culture medium. The project also envisages a comprehensive analysis of the metabolism of this strain for the profile of fermentation products in the presence of various sources of carbon, nitrogen, protein hydrolysates and additional substances as well as their concentrations in the culture medium. In addition, these studies will enable the development of genetic modification methods of the strain under investigation to eliminate or reduce undesirable fermentation products in favor of increasing the level of succinic acid biosynthesis. This will allow to obtain a producer with characteristics comparable to the currently best available strains-producers of succinic acid. The results obtained during the proposed project can provide much new information about the effect of medium components on the regulation mechanisms of fermentation product biosynthesis.