Studies on the prevalence and properties of peptidoglycan peptidases as potential antimicrobial agents with targeted specificity against clinically relevant Gram-positive pathogens

The bacteria of the genera *Staphylococcus*, *Streptococcus* and *Enterococcus* are annually listed as priority pathogens by the World Health Organization in the context of their alarmingly increasing antibiotic resistance. By 2050, an estimated 10 million deaths are projected to occur due to antibiotic resistance of microorganisms, while antimicrobial resistance will cost the world economy $ 100 trillion over the same period. Challenged by these facts, the search for new substances with antibiotic properties is one of the priorities of modern science. Treatment of bacterial infections requires the use of antibiotics. Unfortunately, these drugs, in addition to eliminating pathogens, kill other bacteria, which manifests itself with unfavorable side effects of such a therapy. The concept underlying this research project addresses these negative properties of antibiotics and aims to search for compounds specifically directed against pathogens.

In the course of our research, we discovered a substance - Lysostaphin Sp222, which specifically kills the pathogen *Staphylococcus aureus*, but is harmless to *S. epidermidis*, a bacterium that is a natural component of healthy human skin microbiota. Lysostaphin Sp222 is an enzyme that degrades peptidoglycan - the main component of the bacterial cell wall. The obtained results became the basis of this project, the aim of which is to identify and characterize other peptidoglycan peptidases (PGPs) with bactericidal properties against pathogens of the genera *Staphylococcus*, *Streptococcus* and *Enterococcus*. Based on in-depth bioinformatics analyses it is planned to select genes encoding PGP and next, using molecular biology techniques, to obtain PGSs in a form of recombinant proteins. Then, using microbiological techniques, their activity against pathogenic bacteria will be determined. PGPs with the most promising parameters will be further optimized for specificity and ability to eliminate biofilm as well as tested for cytotoxicity to human cells. These will allow the production of enzybiotics that will meet the overarching principles of novelty and specificity against clinically significant Gram-positive pathogens. This approach fits into the current trend of searching for new solutions for the treatment of bacterial infections in the face of the growing antibiotic resistance of these microorganisms.