Considering that diabetes mellitus is considered as the main risk factor for the development of foot infection, the increasing global population of diabetics is a serious problem threatening the health care systems around the world. It is estimated that half of patients with diabetic foot ulcer develop an infection defined by a common term diabetic foot infection (DFI) which raises serious health consequences, such as increased need for hospitalization, generally low quality of life, high risk of lower limb amputation or even increased risk of mortality. Diagnosis and selection of effective antibiotic therapy of DFI is now a major challenge for both clinicians and scientists around the world due to the wide spectrum of potential pathogens involved in the development of DFI, a long list of potential patient-health-related factors, often polymicrobial nature of infection and increased incidence of drug-resistant bacteria. Literature reports often indicate low efficacy of DFI antibiotics treatment resulting from poor diagnosis of a pathogen. The conventional and molecular techniques of microorganisms identification used so far most often do not allow to take optimal therapy resulted from many factors, such as narrowed spectrum of identified species, difficulty in differentiation of closely related strains or limited ability to detect antibiotic resistance. It is believed that the solution to these problems may be the use of a matrix-assisted laser coupled with mass spectrometry with a time-of-flight analyzer (MALDI-TOF/MS) which has not yet found a wider application in the monitoring and control of diabetic foot infections. Therefore, the main objective of planned research is to adapt the MALDI-TOF/MS technique for quick diagnosis of DFI by creating new protocols and analytical procedures for all stages of the DFI diagnosis - from sampling, transporting and storing clinical samples to microorganisms culturing, their identification and antibiotic resistance detection.

To accomplish the assumed research goal, the following tests will be carried out: (1) determination of culture conditions for microorganisms related to the DFI etiology using reference microbial strains and selection of microbiological media dedicated to various groups of microorganisms; (2) selection of various parameters of sample preparation procedures and MALDI-TOF/MS analysis conditions in terms of their impact on the effectiveness of the reference strains identification in whole-cell and protein extraction mode regarding different types of matrices, cellular extracts preparation methods and antibiotic resistance markers; (3) development of protocols for the procedure of microorganisms identification using molecular biology techniques as a reference method using polymerase chain reaction (PCR) as well as DNA sequencing of different bacterial genome regions including drug resistance genes; (4) investigation of the impact of the method of collection, transport and storage of samples derived from patients suffering from DFI on the final results of microorganisms identification as well as (5) analysis of the microbiological composition of samples derived from different groups of patients using the established protocols for MALDI-TOF/MS and reference molecular methods considering the assessment of the impact of patients' health parameters and antibiotic therapy.

The creation of a new analytical procedures will contribute to the broadening of knowledge about the microbiological background of the diabetic foot infection development including the contribution of drug-resistant strains, as well as expanding the knowledge on the utility of the MALDI-TOF/MS technique as a tool for rapid DFI diagnosis, also in terms of routine clinical laboratories.