

The aim of the project

Antimicrobials are used for many years to treat bacterial infections in humans, in both hospital and community sectors, which resulted in the emergence and persistence of antimicrobial resistance (AMR) within clinical and environmental pathogens. Unmetabolized by living organisms antibiotics, their transformation products, as well as antibiotic resistant bacteria (ARB) and antibiotic resistance genes (ARGs) enter hospital wastewater, which, without being pretreated in hospital wastewater treatment plants, can be transferred to municipal wastewater treatment plants (WWTPs). It is assumed that hospital wastewater discharged to WWTPs is the main anthropogenic source of antibiotics, ARB and ARGs in municipal wastewater, which are then released together with treated wastewater into surface water reservoirs and other elements of the environment. Among the diverse pool of drugs, ARB and ARGs, there are so-called reserve drugs, the priority pathogens with critically high resistance to reserve antibiotics and genes encoding this resistance. Poland is one of the largest consumers of antibacterial drugs in Europe (sixth place among the European Union countries) with an increasing percentage of the appearance of priority pathogens. Many countries of the European Union and beyond, implemented the monitoring of the presence and abundance of the above-mentioned pollutants in wastewater. The study of antimicrobial resistance in hospital wastewater samples is carried out also in Poland, but on a local, not a national level and usually it does not cover the issue of resistance to reserve antimicrobials. **Therefore, the main goal of the project is to include Poland in extensive research on the spread of the resistance to reserve antimicrobials. Hence, the assumption of the project is to create an interactive platform with geographical mapping of risks related to the discharge of the above-mentioned pollutants with hospital wastewater. As a final result, the research can be used to clarify the requirements for hospitals and other health care facilities regarding the use of disinfection of wastewater discharged from individual facilities, as well as the requirements for the microbiological quality of wastewater entering the WWTPs.** The study will be determined on the national level by transdisciplinary team of professionals.

Materials and Methods

To the study, approximately 80-100 hospitals located in various provinces of Poland will be selected differing in terms of the number of hospital beds, the number of patients served and the type of hospital wards. We will determine: basic physicochemical parameters, reserve antimicrobials concentration, critical priority pathogens occurrence, prevalence of genes responsible for resistance to reserve antibiotics in selected isolates, prevalence of genes encoding resistance to reserve antibiotics in hospital wastewater DNA and microbial diversity of sewage microbiome for the presence of priority pathogens. Both standard and advanced techniques will be used for the research, such as the cultivation of priority pathogens on chromogenic media with the supplementation of appropriate reserve antibiotics, nanopore sequencing, quantitative PCR (qPCR) and digital PCR (dPCR) analysis enabling precise, highly sensitive quantification of PCR nucleic acids. The obtained results of studies will be subjected to statistical and bioinformatics analyses.

Results

The project will be used to: (I) generate a new knowledge and demonstrate the distribution of reserve antimicrobials, critical priority pathogens and genes related to resistance to reserve antibiotics in hospital wastewater on national level in Poland, (II) determine the concentration of reserve antimicrobials and the abundance of priority pathogens and genes related to reserve antibiotics in wastewater depending on: the location of hospitals in Poland, the number of hospital beds and the number of patients served, and the type of hospital wards, (III) determine the diversity of reserve antimicrobials, critical priority pathogens and genes associated with resistance to reserve antibiotics in hospital wastewater samples, (IV) assess the health risk ranking of ARGs (associated with human diseases) appearing in the resistome based on metagenomics approach. The obtained results on the frequency of these microorganisms in hospital wastewater will be compared with the frequency of the occurrence of the incidence of diseases caused by these microorganisms recorded by hospitals and units Sanitary and Epidemiological stations operating in a given region. Moreover, the obtained results will be also used to create an interactive new generation web platform, available to all readers, for in-depth analysis and visualization of collected data on AMR. The platform will be used as a tool to inform the society about the presence of antibiotics, antibiotic-resistant bacteria and antibiotic resistance genes, with particular emphasis on reserve drugs, priority pathogens and genes related to resistance to reserve drugs.