## **Description for the general public**

Bacteria are the most abundant and oldest forms of life on Earth which, thanks to their incredible metabolic diversity, can colonize practically all the available niches in the biosphere. They are recognized as the key drivers of biogeochemical processes and produce about half of Earth's oxygen. The leading causes of bacterial mortality are their viruses, bacteriophages (phages). It is estimated that as a result of viral infections 20-40% of the entire bacterial population is lysed every day. Bacterial cell lysis leads to the release of organic carbon and other nutrients back into the environment. Hence, phages can have a significant impact on the microbial food web by increasing the death rates and/or by decreasing growth rates at all trophic levels. Thus, it has become apparent that viruses are a major force behind biogeochemical cycles, and a key element shaping the structure and diversity of microbial communities. Apart from affecting the nutrient cycle, each viral infection may introduce new genetic information into an organism or progeny virus, thereby driving the evolution of both host and viral assemblages.

Bacteriophages are recognized not only as the most abundant but also the most diverse group of biological entitles on Earth. It is estimated that less than 1% of the extant viral diversity has been explored so far e.g. the number of complete phage genomes gathered currently in publicly available GenBank database is not much more than two thousand. Recent advances in DNA sequencing, and especially in the next-generation sequencing technology and bioinformatics tools for analyzing its results (metagenomic tools) have provided novel insights into the ecology of microorganisms found in environmental samples and their diversity. Moreover, metagenomics has become the only currently available approach for studying diverse ecosystems, enabling identification and characterization of unculturable microorganisms. As a result, we learned a lot about viral communities from the largest biomes - marine, soil, freshwater and human-associated. In contrast, relatively little is known about viral populations present in atypical, isolated and extreme (in terms of physical or chemical parameters) environments that are often inhabited by endemic species adapted to such conditions. Our project aims to fill this knowledge gap, at least for a relatively cold and heavy metal-contaminated ecosystem that we selected for our analysis i.e. an abandon for more than 50 years ancient gold and arsenic mine in Zloty Stok (SW Poland). Deadly, for most living organisms, concentration of toxic elements shaped a specific environment that have been inhabited by unique groups of psychrophilic and metallotolerant microorganisms. Many of them form structurally organized communities - mats in the bottom sediments and thick slimy biofilms on the mine walls. These microorganisms, not only grow luxuriate but also act as a biofilters of mine dewatering systems. Although in recent years, this extraordinary ecosystem has been the subject of many studies (in particular due to its application potential), an important element of this niche has been previously overlooked i.e. viruses infecting microorganisms inhabiting this environment. Therefor the aim of this project is to characterize the pool of bacteriophages infecting heavy metal hyper tolerant bacteria inhabiting the heavy-metal-rich Zloty Stok mine. Thanks to that, our knowledge about this ecosystem will expand considerably. In particular, we will focus on viral abundance and diversity. We also would like to estimate the extent of involvement of the mine virome in the shaping and adaptation of the local bacterial community and its involvement in the formation of the spatial structure of rock biofilms and microbiological mat filters.

Although bacteriophages are undoubtedly killers of bacteria they can also act as natural vectors transferring and providing their hosts genes encoding phenotypic traits crucial for adaptation to the various ecological niches e.g. resistance to antibiotics or heavy metals, the ability to biofilm formation or the production of virulence factors. We are going to find out whether such phenomenon also occurs in the microbicenosis of the studied gold and arsenic Zloty Stok mine.

Our research goal will be performed by application two complementary approaches: global (metavirome) and small-scale (the classical analysis of culturable phage isolates). High-throughput sequencing and metagenomic analysis will provide an insight into the viral abundance and diversity within the mine ecosystem, enabling also the discovery of novel viral genomes, and defining the pangenome (total reservoir of genetic information) of the local bacteriophage population. In turn, analyses of the individually isolated culturable bacteriophages will not only help to verify the predictions obtained by the metagenomics approach, but also provide an opportunity to identify, as well as structurally and functionally characterize the complete genomic sequences of bacteriophages directly linked with their specific hosts, including completely novel elements. We assume that our bioinformatic and experimental strategy will lead to the identification of potential parasites of the bacteria, inhabiting this unique ecosystem of the Zloty Stok mine and consequently to the discovery of novel environmental host-phage systems.