microRNAs (miRNAs) are mysterious, small non-coding RNA molecules involved in the regulation of genes expression. They are common in the world of multicellular organism (humans, animals, plants) where play a crucial role in the regulation of important biological processes.

In recent years a lot of number of microRNA sequences were discovered in the genomes of viruses attacking eukaryotic cells, especially herpesviruses. Like all viruses, herpesviruses are parasites and do not have their own metabolism. Herpesviruses belong to the group of large, enveloped viruses, capable of both lytic infection and life-long latency. Within the last decade, it has become obvious that these parasites may use miRNA molecules not only in the switch between life cycles, but also in the regulation of many different metabolic pathways of host to facilitate the optimal production of virus progeny or survival in the conquered eukaryotic cell.

Strong dependence on the host metabolic apparatus is also common among all bacterial viruses, known as bacteriophages (phages). Some of them can play crucial role in the process of genetic variability of the host, leading to the selection of new virulent bacterial strains that can be exemplified by Shiga toxin-producing *Escherichia coli* (STEC). The STEC strains acquired genes coding for Shiga toxins by lysogenization with temperate Shiga toxin-converting bacteriophages (Stx phages). Interestingly, Stx phages and herpesviruses are evolutionary related and show similarities in their life cycles. Moreover, Stx viruses also rely heavily on *E. coli* metabolism after host infection. Stx phages may reside in bacteria in the latent stage of prophage, during which the viral genome is replicated together with bacterial DNA, majority of phage genes are silenced and no new virions are formed. This phase of cycle is named lysogeny and ensures the phage survival in host bacteria for many generations. However, some stress conditions may lead to excision of prophage from host genome and initiation of phage lytic development. During this cycle, Stx virus reprograms host metabolism to provide increased pools of free nucleotides necessary for efficient viral genome replication as well as increased amino acid production for rapid virion assembly what in consequence leads to synthesis of a large amounts of Shiga toxins dangerous to human health and creation of new phage progeny. After bacterial cell death Shiga toxins are released to intestine and then they attack eukaryotic cells. The consequences of this process are difficult to stop bloody diarrhea and human health complication associated with kidney failure.

Reasons for attempting a particular research topic: Importantly, in the literature there are low number reports describing in details how Stx phages alter cellular metabolism and what molecular factors are involved in this process. Perhaps, the key to solving this puzzle are mysterious small, phage-derived RNA molecules. Explanation of this phenomenon will not be easy, because up to now only few very small RNA particles has been reported in prokaryotic world. However, in 2015 our group identified the first functional phage-encoded micro-size RNA molecule, named 24B_1, which probably stimulate indirectly the lysogenic development of Stx phage in bacterial cells. This discovery was a breakthrough not only for us, but also for the whole scientific world, because some researchers deny the existence of such particles in prokaryotic systems.

Project goal: So, in the frame of this project we are going to investigate the biological significance of six newly-identified, phage-encoded small micro-size RNA molecules and explain how they induce metabolic rewiring in *E. coli* bacteria infected with Stx virus.

Description of research: The found molecules are located in the antirepressor region of Stx phage genome and probably take part in the regulation of the switch between life cycles of Stx virus. Essentially, the influence of one of these molecules, named Uproi1, on bacterial amino acid metabolism is very likely. Therefore, we are planning to conduct analysis that allow us to identify metabolic pathways of host cells that are regulated by the phage-derived small RNAs and also characterize the main transcripts of their action.

Substantial results expected: The impact of the proposed project on the development of science and civilization is based on the possibility of discovery of currently unknown regulatory mechanisms of bacterial metabolism that depends on the phage miRNA molecules. Such new information may lead us to understand the diversity of regulation of biological processes which occur during Stx bacteriophages' life cycles in more detail. Moreover, the identification of metabolic targets for newly-identified miRNA molecules will be essential not only in the understanding how Stx viruses conquer their host cells but also may shed a new light on designing of prevention and treatment strategies of STEC infections.