DESCRIPTION FOR THE GENERAL PUBLIC

The term "small molecules with great possibilities" fits the short, about 20 nucleotides in length, RNA molecules, called microRNAs. Since the first such molecule was identified some 25 years ago, in 1993, a growing number of microRNAs has been found in humans, plants and animals. Eukaryotic microRNAs are produced from their own longer precursors and are able to regulate expression of many genes, thereby affecting many cellular processes. Until recently, it was assumed that typical microRNA molecules do not occur in bacterial cell. Bacterial RNA fragments around 100 nucleotides in length have been considered as the most related to eukaryotic microRNAs, however, there is a common opinion that in contrast to eukaryotic microRNAs, they are not processed to shorter forms. Although a few microRNA-size small RNA fragments (15-28 nucleotides in length) have been reported recently in bacteria, these molecules have received little attention up to now.

Recently, we have discovered the first phage-derived, short microRNA size (20 nt) molecule. It was called 24B_1, and this RNA, as well as its longer precursor transcript, were detected in *Escherichia coli* culture after induction of Shiga toxin-converting bacteriophage Φ24B. Phages from this group are responsible for virulence of entrohemorrhagic *E. coli* strains (EHEC). These pathogens can cause serious food poisoning with bloody diarrhea in humans. To our knowledge, the molecule identified by us is the first such small RNA fragment isolated from Shiga toxin-converting bacteriophages which biological significance during phage lifecycle has been determined. As the discovery of both forms: short, 20-nt long and the hairpin-shaped (80-nt long) precursor resemble formation of microRNAs in eukaryotic cells, we suggest that 24B_1 and other analogous molecules (if found) might be formally considered as typical microRNAs molecules in prokaryotic systems. This is important discovery, but in comparison with a huge number of reports concerning eukaryotic microRNAs, we have to realize that current knowledge about such molecules in prokaryotic systems is only "a drop in the ocean", and there is urgent need to develop research in this field. Therefore, in this project we are going to conduct research which will help us to understand how such molecules are formed, what are mechanisms of their actions, and if there are other molecules, similar to 24B_1, affecting the phage development in *E. coli*.

The importance and impact of the proposed project on the development of the research field and scientific discipline is based on the possibility of discovery of currently unknown mechanisms which might revolutionize present view on small bacterial RNAs. This knowledge may provide new insights into the role of microRNA-size molecules during bacteriophage and bacterial development. The scientific importance is additionally increased due to the fact that Shiga toxin-converting bacteriophages and their hosts, entrohemorrhagic *E. coli*, analyzed in this project, belong to dangerous human pathogens.