Lung cancer is one of the most common and deadly type of cancers in the world. The 5-year survival rate of non-small cell lung cancer (NSCLC) has improved significantly, among other things due to the wide access to modern biological therapies, e.g., using anti-PD-1 or anti-PD-L1 antibodies. Unfortunately, many patients whose cancer cells express, the PD-L1 protein do not respond to this type of immunotherapy. Small cell lung cancers (SCLC) constitute a minority of detected cases, but their treatment methods are even less advanced. Considering the above ineffectiveness of therapy, the new diagnostic methods and potential therapies for early cancers detection and monitoring of metastatic changes, are sought. A modern approach is theranostics - diagnostics combined with targeted therapy. The goal of this Project is developing and testing a multifunctional biosensing system for effective lung cancer diagnostics, demonstrating simultaneous therapeutic potential. This system is based on the detection of specific miRNA molecules found in cancer cells – oncomiRNAs, which show a stimulating effect on the progression and invasion of lung cancers, e.g., miRNA-20a and miRNA-661.
For this purpose, we will design two complementary oligonucleotide probes (AntagomiR Molecular Beacon, ARMB) for the above-mentioned oncomiRNAs, labeled with the fluorescent dye and quencher at the end of the sequence. As a result of hybridization of complementary nucleotides probe with targeted oncomiRNA, the probe changes its conformation causing an increase in the fluorescent signal. The emission spectrum of both probes does not overlap, therefore, a single biosensing system can measure the intensity of fluorescence signals of oncomiRNAs simultaneously. This introduces a new, multifunctional dimension of research, because one miRNA is responsible for the increased expression of the PD-L1 protein in lung cancer cells, while the other is a biomarker of prometastatic changes in cancers and its presence may indicate increased invasiveness and aggressiveness of the cancer. The prepared biosensing platform has also high therapeutic potential. Hybridization of the probe with selected oncomiRNAs in lung cancer cells may inhibit the biological activity of oncomiRNAs, potentially contributing to the extinction of their biological activity, by reduction of the PD-L1 or epithelial-mesenchymal transition (EMT) proteins expression. This approach is particularly interesting in the context of the frequent insensitivity to biological treatment and will deepen knowledge in the field of oncogenic cellular processes, which may be closely related to the concentration of oncomiRNA in lung cancer cells. For this purpose, in addition to the standard methods of fluorescence spectroscopy and fluorescence confocal microscopy, we will use cellular functional tests and automated capillary protein electrophoresis, which will allow for insight into molecular changes occurring as a result of the ARMB probe uptake in cells. The feasibility of the developed biosensing system will be investigated with synthesized oligonucleotides in buffer as well as in lysates and healthy and cancer lung cells.