Lung cancer is the cause of the highest number of deaths among patients with malignant tumours. The five-year survival rate is estimated at 15%. The high mortality of lung cancer patients results from the fact that they are often diagnosed at advanced stages of the disease (because of the asymptomatic course of the early stages of the disease), what makes application of the surgically resection impossible. According to the World Health Organization guidelines lung cancer is classified into two subtypes: non-small cell lung cancer (NSCLC) and small-cell lung cancer (SCLC). The studies planned in our project are dedicated the NSCLC since this is the commonest type of lung cancer, as it is diagnosed in approximately 80-85% of lung cancer patients.

There are different treatment options for lung cancer patients. The conventional chemotherapy and radiation were the most frequently options chosen until recently. The discovery of genetic alternations in genes involved in lung cancer pathogenesis, such as epidermal growth factor receptor (*EGFR*), has led to the development of targeted therapies which are currently used as the standard treatment. The latest medical achievement in the treatment of lung cancer is immunotherapy directed at immunoregulatory molecules. Immunotherapy is aimed at intensification of naturally occurring processes involved in the fight against cancer, since it turns out, that some tumours can avoid the response of our immune system by using immune checkpoints – receptors (both inhibitory and activating) which together with their ligands significantly control the immune response.

PD-1 inhibitors are already used in the treatment of some types of tumours, while studies on the use of other immune checkpoints are undergoing intensive research. Our project also deals with this topic. One of the goals of the proposed research is to investigate mRNA and protein expression of PD1, BTLA-4 LAG-3, TIM-3 and TIGIT in non-small cell lung cancer. We would like to establish also the potential reasons for different expression of investigated molecules in tumour tissue. Methylation of regulatory regions of genes is one of the mechanisms which may be responsible for altered expression of given gene. Literature data as well as *in silico* analysis suggest that in genes encoding molecules that are the goal of our research occur regions to which methyl groups can bind what in consequence leads to gene silencing and decreasing of it expression level. Therefore one of the tasks of our project will be checking if investigated by us genes are methylated in tumour tissue and if the methylation status is correlated with their expression level. Altered expression of gene may be also caused by miRNAS. This molecules binds to specific sequence especially in 3'untranslated region (3'UTR) of gene leading to downregulation of its expression. In our project we plan to investigate the expression of selected miRNAs and correlate the level of their expression with expression of investigated in our project molecules. Moreover we will screen some regions of investigated by us genes in search for potentially functional single nucleotide variants capable to affect their expression by alternating binding sites for miRNA and transcription factors. If we find such variants we will try to evaluate their functional values.

Despite the intensity of conducted research the role of new immune checkpoints such as BTLA, LAG-3, TIM-3 and TIGIT as well as the role of mechanisms regulating their expression is still not well elucidated and understand in the context of NSCLC. In proposed research we would like to investigate the expression of these molecules in NSCLC tissues as well as study the genetic and epigenetic mechanisms involved in regulation of their expression. We anticipate that the results obtained from proposed research may not only expand up-to date knowledge about these immune checkpoints but also may have clinical implication – contribute to establishing prognostic factors for better selection of patients to immunotherapy and the may have also implication for the selection of the most appropriate treatment options for individual patients (e.g. consideration of including of demethylating agents to the treatment protocol).