Immunosuppressants are important drugs used in autoimmune diseases treatment and after organ transplantation to minimize the risk of the transplant rejection. Thiopurines, are one of the examples of this class of drugs. Azathioprine is a thiopurine-type commonly used prodrug, which is converted in the liver and kidneys to drugs as 6-mercaptopurine and 6-thioguanine. The activity of thiopurines relays on the inhibition of DNA synthesis in cells. In these DNA molecules regular purines (guanine and adenine) are replaced by thiopurines, which disables DNA replication, inhibiting proliferation of cells involved in the immune response. The therapeutic effect of azathioprine occurs after weeks or months of medication. Unfortunately, long-term administration of azathioprine can lead to many serious side effects such as nausea, vomiting, diarrhea, fatigue, acute pancreatitis (especially in patients with Crohn's disease), anemia and reduced resistance to infections. To select an effective dose, while minimizing side effects, the patient must undergo regular tests. Currently, the most popular methods rely on the determining i) the thiopurines active metabolites - thioguanine nucleotides (TGN) in red blood cells or ii) the number of leukocytes. Unfortunately, both indicators are affected by additional factors, such as viral infection, therefore it is not always possible to obtain reliable results. Therefore, there is a need to develop new methods to control thiopurine levels in body fluids. As already mentioned, the mechanism of thiopurines action is their interaction with DNA, however, the nature of these interactions is not fully understood. The scientific problem undertaken in this project is to examine and describe the interactions occurring between the DNA strand, that is the proposed bioreceptor and immunosuppressive thiopurine analogues. The comprehensive description of these interactions will be the first step in developing an effective method of thiopurine determination. To accomplish this task, it is necessary to propose a measuring system with properties adapted to the analyzed substances. In this project gold screen-printed electrodes, modified with gold nanoparticles, to which DNA fragments will be attached by linkers will be used. Gold nanoparticles significantly increase the electrode active surface and enhance the electron transfer between the bioreceptor and the electrode, which in turn increases the sensitivity of the measuring system. Moreover, the selected bioreceptor can be easily attached to the surface of nanoparticles. Typically, bifunctional thiols are used, whose thiol group binds to the gold surface, and the second functional group serves as a bonding site for bioreceptors immobilization. However, the number of receptors connected with the surface is relatively small. To increase the amount of surface-connected bioreceptors, we propose the alignment of hyperbranched molecules – dendrimers. The name of dendrimer comes from the Greek word, which in translation means *tree*, and it excellent reflects its structure. Dendrimers consist of repetitive connections of organic compounds that split like branches from the tree bough. Thanks to this branched structure, there may be dozens of functional groups on the dendrimer surface, which can be used to immobilize the bioreceptor. The number of receptor molecules connected with the electrode surface has a significant impact on the recorded signal intensity. The proposed electrodes modified with the nanogold-dendrimer-DNA system will assure the sensitive and selective measuring system for study on the DNA-thiopurines interactions. These studies will give base for the description of DNA-thiopurines interaction and may also shed light on the cause of the azathioprine side-effects. Obtained information will be vital for further investigation on the development of selective biosensors for thiopurines detection. This will open the door for manufacturing of the disposable sensors, that can be used in clinical practice to monitor the concentration of these drugs in patients' blood.

The research intentions presented in the project are fully in line with current global trends in materials science research related to the preparation of hybrid bioplatforms, which can be used in real-time medical diagnostics performed at the patient's bed (co called Point of Care system).