Programmed cell death is needed in normal development, homeostasis and to prevent abnormal proliferation diseases, such as cancer. One of them, recently discovered, is ferroptosis, regulated cell death depending on iron. Morphological features of ferroptosis include intact cell membrane devoid of blebbing, normal sized nucleus free of chromatin condensation and dense miniature mitochondria, with vestigial cristae. Biochemically, ferroptosis is characterised by accumulation of Reactive Oxygen Species (ROS), and lipid peroxidation products. Normally, in the cell many anti oxidative processes take place, both enzymatic and non-enzymatic, to prevent massive lipid peroxidation. But in the case of insufficient amount of antioxidants, the imbalance in the rate of ROS generation and detoxification occurs. Such state is called oxidative stress. In ferroptosis, oxidative stress is caused by the inhibition of an antioxidant enzyme GPX4, and this state can be achieved by inhibition of the cysteine-glutamate antiporter System Xc-. Elevated level of iron inside the cell leads to Fenton reaction, in which free radicals are formed. This two processes, i.e. ROS overproduction and insufficient antioxidant mechanism, together provide to DNA, proteins and lipids damage. The most characteristic feature of ferroptosis is uncontrolled oxidation of polyunsaturated fatty acids (PUFAs) and generation of fatty acid radicals.

Since the name ferroptosis was introduced in 2012, many researches was done to identify possible mechanisms of its induction and inhibition. One of them is relation of iron transport via transferrin (Tf) and transferrin receptor-1 (TfR1) system with heat shock protein B1.. During the process of such iron import, the Tf-TfR1 complex is formed. The TfR1 receptor binds two Tf, with two iron Fe(III) ions bound by each. Tf-TfR1 undergoes internalisation by endosome formation. Then, inside the endosome, pH change cause release of Fe(III) ions, which are subsequently reduced to Fe(II) and in such oxidation state exported outside endoscope. TfR1 is recycled to cell membrane and Tf released outside the cell. It was shown, that a member of small heat shock proteins, heat shock protein-B1 (HSPB1), is able to inhibit the recycling of TfR1 and, despite increased TfR1 expression, lower the iron level inside the cell. The proposed mechanism include the rigid cortical actin cytoskeleton, which acts as a barrier blocking off moving of the TfR1-Tf containing cytoplasmic vesicles to the cell surface. But, after a thorough search of the PDB database, I found in the amino-acid sequence of HSPB1, the same motif as in ferritin (an iron storage protein). This fact allows to put a hypothesis, that iron level reduction may occur not only by blocking the iron import, but also by direct Fe(II) or Fe(III) ion binding by HSPB1. To check, if this is true, we have selected several peptide fragments from the structure of HSPB1, and we will provide physico-chemical experiments, to determine thermodynamic parameters of complexes of Fe(II) and Fe(III) with the chosen peptides, stability of formed complexes, their geometry, and indicate potential binding sites for Fe(II) and Fe(III) ions. Furthermore, we will define the red-ox properties of studied complexes, to indicate, whether they could be reduced/oxidized at physiological conditions, and take part in ROS formation. The similarity of the potential iron binding motif from HSPB1 with confirmed iron binding motif from heavy chain of ferritin is interesting, because this ferritin chain is responsible for the reuse of iron. Therefore, iron binding by HSPB1 might be „temporary", and changes of conditions inside the cell might result in iron ions release.

The interesting fact about iron and HSPB1 is that elevated levels of both of them are found in cancer cells.. HSPB1 was identified as a critical regulator of cancer cell death, because due to high iron and HSPB1 levels, the cells might be predisposed to ferroptosis if the HSPB1 pathway is down-regulated. It opens the possibility of combinational cancer therapy, which should target both HSPB1 and ferroptosis. Additionally, some diseases are connected with pathogenic role of ferroptosis, such as Huntington's disease, acute renal failure or periventricular leukomalacia. It was proven, that in these diseases HSPB1 has a protective role. Hence, elucidation of potential mechanisms of HSPB1-mediated ferroptosis inhibition under various conditions is needed.

As a result of the project we want to explain, if the interaction of HSPB1 is possible, because it will be important in elucidation of the mechanism of ferroptosis inhibition by HSPB1. Therefore, the results of proposed studies will contribute to expanding the bioinorganic chemistry of HSPB1 interactions with Fe(III) and Fe(II), what will be a valuable basic knowledge, crucial before any potential ferroptosis-dependent diseases and cancer treatment strategies can be designed.