DOCTORAL PROJECT PROPOSAL
DOCTORAL SCHOOL OF EXACT AND NATURAL SCIENCES
NICOLAUS COPERNICUS UNIVERSITY IN TORUŃ
Grant Fellowship, August 2020

Project title (in English)		
Computational studies of regulatory mechanism and inhibition of ferroptotic cell death signal.		
Project title (in Polish)		
Komputerowe badania mechanimu regulacji i hamowania sygnału śmierci komórki na drodze ferroptozy.		
Project submitter		
Dr Karolina Mikulska-Rumińska degree/title, first and last name	karolamik@fizyka.umk.pl e-mail address	
	WFAiIS UMK organizational unit	
Suggested supervisors and mentors		
1) main supervisor		
Prof. dr hab. Wiesław Nowak degree/title, first and last name	wiesiek@fizyka.umk.pl e-mail address	
	WFAiIS UMK organizational unit	
	field:	Physics/Biophysics
2) auxiliary supervisor		
Dr Karolina Mikulska-Rumińska degree/title, first and last name	karolamik@fizyka.umk.pl e-mail address	
	WFAiIS UMK organizational unit	
	field:	Biophysics

1. PHD PROJECT DESCRIPTION (4000 characters max., including the aims and work plan)

Project title:

1.1. Project goals

- To characterize the regulatory mechanisms triggering the activity of the PEBP1-15LOX protein complex and roles of other components in the ferroptosis process.
- To identify ligands diffusion pathways in 15LOX and PEBP1-15LOX.
- To develop *in silico* new inhibitors which will stop ferroptotic cell death signal initiated by the PEBP1-15LOX complexation.

1.2. Outline

Ferroptosis is new, recently recognized, non-apoptotic form of cell death characterized by the accumulation of lipid peroxides. Is plays a vital role in the treatment of cancers, and can contribute to the degradation of tissue in brain trauma, kidney disease and asthma. Currently, over 365 million people globally are affected by asthma and there is no effective prevention for it. Better insight into ferroptosis may be new hope to them.

In SONATA research project, funded by NCN, we would like to verify hypothesis that the understanding of mechanisms of ferroptosis process at the molecular level may bring novel chemical compounds acting against the ferroptotic cell death. Our recent studies (S. Wenzel et. al., *Cell*, 2017) showed that formation of PEBP1 with 15LOX protein complex initiate the ferroptotic cell death signal i.e. leads to lipids peroxidation. This discovery sparked substantial interest in research community, despite the fact that molecular mechanism of this association in poorly understand yet. Thus, we would like to fulfill this gap and bring new knowledge about 15LOX-PEBP1 complex dynamics and its functionally important regions using multiple advanced computational approaches.

The main goal of present research project is to develop *in silico* new inhibitors which will protect a cell against ferroptotic death signal activated by the PEBP1-15LOX complexation, which in turn is an initial step of lipids peroxidation. Such inhibitors may potentially lead to new drugs developments. The results obtained during this project will have significant impact on the biomedical field. Promising outcome will be shared with the US collaborators for biochemical verification of our computational predictions.

1.3. Work plan

1) Mastering molecular dynamics, ligand parametrization, molecular docking and Python programing skills.

2) Finding a general approach for recently developed method based on the enhance sampling (implemented in PLUMED) to reveal new entries and tunnels in isolated 15LOX protein and 15LOX complexed with PEBP1.

3) Finding hot spots and crucial sites for allosteric modulation and dynamics using available and/or novel computational tools and analysis methods.

4) Mastering pharmacophore model preparation and usage of virtual screening.

5) Based on knowledge from tasks #2-4 providing a new set of potential inhibitors for 15LOX and 15LOX-PEBP1 complex.

6) The most promising compounds will be tested in biochemical studies by the US collaborators.

1.4. Literature

1. Kapralov, Q. Yang, H. Dar, Y. Tyurina, T. Anthonymuthu, R. Kim, C. Croix, **K. Mikulska-Ruminska**, et al., *Redox Lipid Reprogramming Commands Susceptibility of Macrophages and Microglia to Ferroptotic Death*, Nature Chem. Biol. 3 (2020) 278-290.
2. **K. Mikulska-Ruminska**, et al., *Characterization of Differential Dynamics, Specificity, and Allostery of Lipoxygenase Family Members,* J. Chem. Inf. Modeling, 59 (2019) 2496-2508.
3. T. Anthonymuthu, E. Kenny, I. Shrivastava, Y. Tyurina, Z. Hier, H. Ting, H. Dar, V. A. Tyurin, A. Nesterova, A. Amoscato, **K. Mikulska-Ruminska** et al., *Empowerment of 15-Lipoxygenase Catalytic Competence in Selective Oxidation of Membrane ETE-PE to Ferroptotic Death Signals*, HpETE-PE. J. Am. Chem. Soc. 140 (2018) 17835-17839.
4. H. Dar, Y. Tyurina, **K. Mikulska-Ruminska**, et al., *Pseudomonas aeruginosa utilizes host polyunsaturated phosphatidylethanolamines to trigger theft-ferroptosis in bronchial epithelium*. J. Clin. Invest. 128 (2018) 4639-4653.
5. S. Wenzel, Y. Tyurina, J. Zhao, C. Croix, G. Mao, V. Tyurin, T. Anthonymuthu, A. Kapralov, **K. Mikulska-Ruminska**, et al., *PEBP1 Wardens Ferroptosis by Enabling Lipoxygenase Generation of Lipid Death Signals,* CELL 171 (2017) 628-641.

1.5. Required initial knowledge and skills of the PhD candidate

- Analytical thinking
- Eager to learn and focus on the goals
- Skills in programming, preferably in Python
- Good understanding of physics
- Basic understanding of biology and/or chemistry
- Involvement in scientific work

1.6. Expected development of the PhD candidate's knowledge and skills

- Improving knowledge in biophysics, bioinformatics and computer science.
- Improving knowledge of protein database services and other computational tools and methods.
- Improving programming skills in Python programming language.
- Improving English proficiency.
- Networking - making new connections with other scientist in Poland and beyond.