

Neutrophils belong to white blood cells and its main function is to defend the organism against pathogens. For this purpose, these cells are equipped with antibacterial agents, which are packed within the granules. These granules contain primarily proteolytic enzymes such as elastase, proteinase 3, and cathepsin G. Neutrophils use three main tactics to eliminate the enemy: the first is degranulation - the secretion of bactericidal agents outside the cell, the second is phagocytosis - the engulfment and intracellular destruction of the pathogen, while the third tactic is netosis. Netosis is the process of DNA secretion from the cell nucleus forming a trap to catch pathogens.

Granzyme A is one of the proteolytic enzymes found in the neutrophil granules. So far its role, activity and exact location in these cells are unknown. The reason for this is low concentration of this enzyme in neutrophils and also technological limitations.

Neutrophils are extremely short-lived white blood cells circulating in human peripheral blood. These cells are terminal, which means that they die several hours after being generated from myelocytes. In order to play their physiological functions neutrophils are easily activated, which on the other hand is a great challenge in laboratory tests. Moreover, isolated neutrophils survive on average about eight hours, making some experiments impossible to perform. The best method to test enzymes is to use genetic engineering techniques and to produce cells without the targeted enzyme. In the case of neutrophils, this can only be done on their progenitors, which can be found in the bone marrow, but ethical issues and painful samples collection are the most important limiting factors. Another possibility for genetic modification of neutrophils for serine proteases tests is the use of mouse models, but these tests are very expensive and require special facilities. Considering the above arguments, research on neutrophils proteases is problematic and, together with the low expression of GrA in these cells, is a big challenge to investigate this enzyme and therefore this topic seems to be neglected.

In our project we propose a new model for research on granzyme A using neutrophils made from iPS cells. From these almost all cells, including neutrophils, can be produced. This model will allow us to make genetically engineered neutrophils without taking the bone marrow samples and will allow for an extensive GrA study. We believe that this strategy will allow us to study the functionality of GrA in neutrophils and indicate their role in protecting the host against pathogens.