Telomeric shortening in the long-term allogeneic hematopoietic cell recipients compared to their donors

Telomeres are short DNA fragments of repeatable sequence which are localized at each chromosome endings. Their primary role is buffering against DNA loss that happens at the chromosomal ends every round of replication - simply providing excess non-coding DNA to be lost. In consequence, after each cellular division (and associated replication), telomeres are shortened. When they reach critical length, cellular divisions are inhibited and cell enters senescence (cell ceases to divide) or apoptosis (cellular suicide). Therefore, telomeric length is cellular senescence indicator. During the process of allogeneic hematopoietic cell transplantation, limited number of stem cells engrafted to the recipient must reconstitute the whole functional hematopoietic system. This requires immense proliferation activity; consequently, multiple rounds of replication have to take place. We assume that such process should result in substantial telomere shortening, thereby in accelerating cellular senescence.

Primary goal of our project is to compare dynamics of hematopoietic cells senescence in the long-term survivors of allogeneic hematopoietic cell transplantation (HCT) and their donors by the measurement of difference in their mean telomeric length in subpopulations of TCD4+ and TCD8+ lymphocytes. After successful transplantation recipient's lymphocyte population originates directly from the transplant donor cells. This allows to measure a postulated difference in telomeric length between cells that were transplanted and the cells that remained in the donor. The analysis will be performed using a Flow FISH method. It utilizes flow cytometry technique, allowing for a single-cell fluorescence analysis based on in situ hybridization of fluorescent, telomere-specific probe. Acquired fluorescence intensity should be proportional to each cell mean telomeric DNA length. After acquisition of the results, we shall determine if there is any difference in recipients mean telomeric length in respect to age of the donor and occurrence of severe chronic graft versus host disease.

The reason we took up the topic of shortening telomeres in the context of hematopoietic cell transplantation was the work of Professor Jan Maciej Zaucha et al. from 2001. They studied the length of telomeres in dog cells which were subjected to unusual proliferative stress through repeated irradiation of the whole body. Blood cells transplanted to allogeneic recipients are also subjected to enormous proliferative stress. Because of the access to a relatively large population of long-term recipients of an allogeneic haematopoietic cell transplant, we decided to carry out similar research in humans using transplantation as expositional proliferative stress to revisit the unanswered question - what happens to the telomere length in cells that were transplanted. We believe that the results we obtain would contribute to further development of knowledge in the field of cellular senescence and aging *per se* and allow to determine potential factors that might influence the process. It will also provide more solid foundations for the key question whether allogeneic hematopoietic cell transplantation accelerates cellular senescence or merely reduces stem cells potential pool of divisions in post transplantation period.