Justyna Korycka

Identyfication of ZDHHC gene products in hematopoietic cells, cancer cells of hematopoietic origin and in epithelial cells. The effect of DHHC proteins on cell cycle of erythroid cells.

SUMMARY

Present literature clearly indicates that palmitoyltransferases belonging to the DHHC family of proteins are essential to normal function of the cell, as they are the enzymes that catalyze palmitoylation, one of the most common post-translational lipid modifications of proteins. It was found that mutations in *ZDHHC* genes which abolish the PAT activity of DHHC proteins lead to serious pathologies. These mutations are connected with certain neuronal and mental disorders such as mental retardation (DHHC9, DHHC15), Huntington's disease (DHHC17), schizophrenia and bipolar disorder (DHHC8), as well as with cancerogenesis (DHHC2, DHHC9, DHHC11, DHHC17). However, little is known what are the effects of mutations in genes coding for DHHC1, DHHC4, DHHC6, DHHC16, DHHC18, DHHC19, DHHC20, DHHC22, DHHC23 and DHHC24 and whether they are related to pathological processes.

So far, little is known about the distribution of each of the 23 human DHHC proteins in cells of individual stages of erythropoiesis. Moreover, the data about expression of *ZDHHC* genes in many other types of cells are also insufficient. For this purpose, new library of oligonucleotide primers for selective amplification of each *ZDHHC* genes transcripts as well as all ("active") isoforms of these genes was constructed. Moreover, control genes (with constantly low and constantly high expression levels) products detection system was selected and primers for detection of "impurities" in specific hematopoietic cells population were designed. Our studies first of all demonstrate which of 23 *ZDHHC* genes are expressed in human erythropoietic cells. During these studies it was shown that DHHC17 is the only PAT gene transcript found in the reticulocyte mRNA. The presence of this gene product was also detected in the erythrocyte membrane by immunoblotting. Furthermore, it was shown that, among hematopoietic cells, DHHC17 is present only in erythropoietic cells and platelets which during hematopoiesis originate from the common precursor named MEP (megakaryocyte/erythrocyte progenitor).

Cells in which the expression level of *ZDHHC* genes was decreased using specific DNAzyme are characterized by cell cycle disturbances. Moreover, morphology of these cells also differ from the control, because they are larger then normal cells. Apart from that, it was found that cells transfected by DNAzyme demonstrate decreased proliferation rate compared to the control.

The next step of this research project was the detection of *ZDHHC* genes transcripts in various, different from erythroid, hematopoietic cells what in consequence allowed to conclude which of the *ZDHHC* genes are expressed in these cells. Furthermore, the expression analysis of genes coding for individual palmitoyltransferases in cells from selected human cancer cell lines derived from all three primary cell layers of animal embryo was undertaken. Thus, as a result of above research, qualitative "profiles" of DHHC enzymes in many different type of cells were obtained.

Taking into account the information available in the literature, regarding the linkage of defects in the genes coding for DHHC enzymes with various pathologies, detection of these genes products may be crucial to study the molecular basis of certain diseases and may contribute to progress in the therapy of these diseases.