Abstract

Photodynamic therapy (PDT) is a treatment modality for cancer. It combines selective accumulation of chemical compounds, called photosensitizers (PS), with light to irreversibly damage cancer cells via oxidative stress. The main goal of this thesis was to study photosensitizers represented by a unique group of newly synthesized porphyrin derivatives with glycol chain substitution. Glycol-functionalized porphyrins containing one to four low molecular weight glycol chains that are linked via ether bonds to the meta-phenyl positions of meso-tetraphenylporphyrin (mTPP(EG)1-4) were compared with fluorinated (pTPPF(EG)4) and nonfluorinated (TPP(EG)4) derivatives having glycol chains in para-phenyl positions. The cellular uptake and photodynamic activity was significantly dependent on terminal groups of the glycol substituent. Hydroxy glycol porphyrins, in contrast with methoxy glycol porphyrins, exhibited efficient intracellular transport and high induction of apoptosis in tumor cell lines in vitro. After initial testing effective prototype hydroxy ethylene glycol derivatives were selected and analyzed in detail. Para derivatives pTPP(EG)4 and pTPPF(EG)4 accumulated mainly in lysosomes whereas meta derivatives mTPP(EG)1-4 in the endoplasmic reticulum (ER). Position of ethylene glycol chain on the porphyrin ring affected not only intracellular localization but also PDT efficacy demonstrated by permanent ablation of human breast carcinoma (MDA-MB-231) in nude mice following treatment with meta derivatives. After photoactivation, both types of derivatives induced death of tumor cells via reactive oxygen species (ROS). Para derivatives pTPP(EG)4 and pTPPF(EG)4 activated the p38 MAP kinase cascade, which in turn induced the mitochondrial apoptotic pathway. In contrast, meta porphyrin derivative mTPP(EG)4 induced dramatic changes in Ca^{2+} homeostasis manifested by Ca^{2+} rise in the cytoplasm, activation of calpains and stress caspase-12 or caspase-4. ER stress developed into unfolded protein response. Immediately after irradiation the PERK pathway was activated through phosphorylation of PERK, eIF2α and induction of transcription factors ATF4 and CHOP, which regulate stress response genes. PERK knockdown and PERK deficiency protected cells against mTPP(EG)4-mediated apoptosis, confirming the causative role of the PERK pathway.

Analysis of the cell-death mechanism revealed that mTPP(EG)4 represent a novel nonmitochondrially localized photosensitizer that has a profound ability to induce apoptosis in tumor cells and exhibits a superior PDT efficacy in elimination of experimental tumors in comparison to clinically used photosensitizer Foscan. The presented work thus suggests an

interesting avenue for further development of photosensitizers aiming at their future clinical application.