ABSTRACTS OF THE CONFERENCE

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G-QUADRUPLEX DNA BINDING AND TELOMERASE INHIBITION BY PHEOPHORBIDES

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Introduction. G-quadruplexes (G4) are specific structures formed by some guanine rich DNA sequences, e.g. those at the ends of the telomeres. Their stabilization by small molecules can lead to telomerase inhibition and thus is a promising anticancer strategy. This research was aimed at the study of DNA binding and telomerase inhibition of a series of pheophorbides, compounds of porphyrin family.

Methods. Fluorescent Intercalator Displacement (FID) method was used to study G4 and duplex DNA binding affinity and selectivity of three pheophorbides: natural anionic Pheophorbide-a and its neutral and cationic derivatives. The assay is based on the substitution of Thiazole Orange dye in fluorescent DNA complex by a ligand resulting in concentration-dependent fluorescence decrease that allows the determination of binding constant and stoichiometry. Quadruplex was formed by Tel22 oligonucleotide d[AGGG(TTAGGG)$_3$]. DNA binding constants (K_b) were obtained from the titration data by Scatchard method. Telomerase inhibition activity of compounds was determined by Telomeric Repeat Amplification Protocol (TRAP) *in vitro* assay.

Results. DC_{50} parameters (ligand concentration required to induce 50% fluorescence decrease) were in the range of 19.5-62.7 and 11.4-37.2 μM for G4 and duplex DNA, respectively. K_b values of compounds were (2.1-5.6)×10^6 M^{-1} for G4 and 7×10^4-3.1×10^6 M^{-1} for duplex DNA. The highest selectivity for G4 vs. dsDNA was observed for cationic derivative Cat-Pheo-*a* (selectivity index 1.9). IC_{50} values for telomerase inhibition were in the range of 8-40 μM.

Discussion. Binding constants indicate high G4 affinity of pheophorbides. Cat-Pheo-*a* demonstrates both high affinity for G4 DNA (DC_{50} 19.5 μM, K_b 4.5×10^6 M^{-1}) and good selectivity to quadruplex DNA. This G4 ligand has also the highest inhibition activity towards telomerase (IC_{50} 8 μM). The highest DNA binding efficiency and biological activity of Cat-Pheo-a can be due to the presence of cationic trimethylammonium group able to form ionic bonds with DNA phosphates.

Conclusions. Cat-Pheo-*a* is the most efficient G4 ligand and telomerase inhibitor among the studied compounds.

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