ABSTRACT

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Title of Diploma Thesis: **Modulation of pK_a of the recognition moiety of azaphtalocyanine sensors**

Azaphthalocyanines (AzaPc) are macrocyclic compounds containing a large system of conjugated double bonds that enables them to absorb light in the red part of the spectrum that is promising in biological applications. They are characterized by intense red fluorescence as one of the relaxation pathways of the excited state after absorbing a photon. The fluorescence of AzaPc substituted with a phenol moieties on the periphery can be switched ON/OFF depending on the pH of the environment and the pK_a of the phenolic group. In basic medium, the molecule occurs as phenolate and undergoes intramolecular charge transfer between the phenolate group (a donor) and the electron-deficient macrocyclic core (an acceptor). As a consequence of this process, the fluorescence is quenched. Switching between ON/OFF states in phenol-substituted AzaPc is dependent on the proton concentration and thus can be utilized in pH sensing.

The aim of this work was to synthesize derivatives of phenol-substituted AzaPcs whose pK_a is modulated with suitable substitution in *ortho* position. The synthesis started with preparation of appropriate precursors (*i.e.* substituted 5-(4-hydroxyphenyl)pyrazine-2,3-dicarbonitriles). Electrophilic substitution of commercially available 4-hydroxyacetophenone (bromination or nitration) was performed. The products were treated with selenium dioxide affording corresponding ketoaldehydes that were not isolated but directly reacted in a condensation reaction with diaminomaleonitrile. To obtain unsymmetrical AzaPc, a mixed cyclotetramerization (statistical condensation) of these precursors (A) with 5,6- bis(*tert*-butylsulfanyl)pyrazine-2,3-dicarbonitrile (B) was performed using magnesium butoxide as an initiator. Resulting mixture contained six different congeners (*i.e.* AAAA, AAAB, AABB, ABAB, ABBB, BBBB) from which the required congener (ABBB) was isolated using column chromatography. AzaPcs were then incorporated to lipophilic particles (microemulsion, liposomes) and the fluorescence changes were investigated as a function of pH of the buffer. Dependence of fluorescence on pH allowed determination of pK_a value.