Effects of DNA Sequence and Topology on the Binding of Porphyrins and Psoralens to DNA using Restriction Enzyme Activity Assays

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Psoralens are DNA-binding agents which can photo-crosslink DNA at AT sequences. The sequence specificity of the equilibrium binding [which precedes crosslinking] of 8-methoxypsoralen and of 5-methoxypsoralen has been examined using restriction enzyme activity assays and preferred binding sequences located on phiX174 DNA. The sequence specificity of N-methylpyridyl porphines for AT-rich sequences has also been examined using restriction enzyme activity assays. The binding behavior of both of these molecule types suggests that they may distort the DNA structure on binding. Thus, the binding of a porphyrin may alter the binding of a psoralen – and visa versa. In this project, the interplay between the porphines and psoralens on binding to phiX174 DNA will be examined using restriction enzyme activity assays employing enzymes which cleave DNA at different sequences. The experiments will be done with a constant 8-methoxypsoralen concentration and a varied porphine concentration; then the experiments will be performed with varied 8-methoxypsoralen and constant porphine. These experiments will provide further information on the nature of the binding of these compounds to DNA and on how they distort DNA structure. Many anti-cancer and anti-microbial agents work by binding to DNA and subsequently altering the structure and functioning of the DNA. Various agents including psoralen and porphyrin have been shown to bind cooperatively to DNAs. Psoralen and porphyrin drug families are DNA-binding agents that attach at AT sequences. The use of both drug compounds alters the structure of DNA, which this project will be studying.