The Synthesis and Biological Evaluation of Zinc (II) DPP Carbohydrate Analogs

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Introduction: Photodynamic therapy (PDT) has increasingly gained attention as an effective treatment for cancer. Esophageal and lung cancer, the two primary forms that PDT is currently approved to target, are expected to kill over 165,000 Americans in 2010 alone, making the improvement of current therapies of vital importance.

PDT makes use of a photosensitizer that, when irradiated with wavelengths of visible or near-infrared light, creates reactive oxygen species (ROS) that can kill target tissues through cell necrosis or apoptosis. Traditionally, porphyrin molecules, with their strong light-absorbing properties, have played an important role in PDT.

A number of simple mono and disaccharide-based carbohydrate-porphyrin conjugates have recently been developed as PDT agents. These compounds show improved therapeutic efficacy over their non-glycosylated counterparts due to their inherent solubility in aqueous solvents, a condition necessary for their introduction into biological fluids, and their enhanced tumor cell selectivity. This selectivity is mediated by the binding of carbohydrates to proteins such as galectin-1, a β-galactoside carbohydrate binding protein overexpressed on tumor cells and which has been implicated in tumor transformation and metastasis. While the use of defined carbohydrate structures to selectively target tumor cells expressing tumor specific carbohydrate-binding proteins shows great potential, this line of research has not yet been fully explored.

One reason for the lack of research in this area may be that many attempts to synthesize carbohydrate-porphyrin conjugates have led to the production of the desired products in low yields. Recently, Snyder and coworkers have developed a palladium-catalyzed cross-coupling strategy as a new route to carbohydrate-porphyrin complexes. Snyder’s methodology has provided consistent access to carbohydrate-porphyrin conjugates in high yields; however, the necessity of being conducted under Schlenk conditions has limited the scalability of this reaction.

In addition, the use of palladium, an expensive and toxic metal, as a catalyst makes the development of a new protocol for the synthesis of these molecules especially attractive.

Hypothesis: Cu(I)-catalyzed azide-alkyne 1,3-dipolar cycloaddition, also known as “click” chemistry, has been employed in the facile synthesis of a number of carbohydrate bioconjugates. Despite this, there has been little done to generate carbohydrate-porphyrin conjugates, which could be used in PDT, via this method. Therefore, the specific goals of this research are two-fold: (i) to design a rapid and facile approach to the synthesis of these conjugates using glycosyl azides and porphyrin alkynes; and (ii) to evaluate the efficacy of these compound as viable PDT agents.

I have chosen to use a zinc-metallated DPP derivative as the underlying porphyrin structure because I feel this will increase the cytotoxicity of the compound. Zinc is known to exhibit anti-tumor properties on its own and diphenyl porphyrins have been shown to possess increased therapeutic activity compared to their corresponding tetraphenyl derivatives. The conjugation of carbohydrates to the central porphyrin ring will increase the compound’s solubility in aqueous solutions. In addition, the use of designed carbohydrate structures, which selectively target carbohydrate binding proteins, can be used to selectively target tumor cells.
Methodology: The first step in this procedure is the synthesis of alkyne substituted diphenyl porphyrin (Scheme 1). The TMSA-substituted benzaldehyde derivative 1, which is readily available from bromobenzaldehyde, will first be converted into the alkyne-substituted DPP derivative 2 using a procedure reported by Lindsey#. TBAF deprotection to yield the terminal alkyne, followed by metallation with zinc, will yield porphyrin 3.

A variety of acetate-protected carbohydrate azides will be conjugated to porphyrin 3 via a Cu(I)-catalyzed azide-alkyne 1,3-dipolar cycloaddition. This, followed by standard acetate deprotection chemistry, will yield the final conjugate 4. Initially, glucose, galactose, and N-acetyl glucose will be evaluated as a proof of concept. Further work will involve the conjugation of lactose, and then N-acetyllactosamine, the minimal ligand for galectin-1.

Scheme 1. The synthesis of glycosylated DPP derivatives.

The final carbohydrate-porphyrin conjugates will be evaluated against a line of HEp-2 carcinoma cells in order to determine their cytotoxicity. The photosensitivity of the compounds will be assessed by the amount of free radicals they produce upon irradiation with specific wavelengths of light, while their ability to bind to galectin-1 will be determined via protein-protein binding assays.

Expected Results: This procedure represents an improved synthetic method for the generation of carbohydrate-porphyrin based PDT photosensitizers. The cytotoxicity of the compounds against cancer cells should also be greater than that of the more common tetraphenyl porphyrin derivatives and those lacking Zn metallation. Due to the strong binding of N-acetyllactosamine to galectin-1, the DPP derivative conjugated to this carbohydrate should represent the most therapeutically effective photosensitizer in the series. Future work will involve the conjugation of more complex carbohydrates to explore how the sugar substituent influences the compound’s efficacy. The effect of differing the underlying porphyrin structure could also be studied.