Short Communication

New Photosensitizers for Photodynamic Therapy: Syntheses of Chlorin e6 Dimer and Trimer

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The hematoporphyrin derivative (HPD) obtained from hematoporphyrin by acetylation and subsequent alkaline treatment is a photosensitizer widely used for the photodynamic therapy of cancer (PDT).¹⁾ Its tumor-localizing components have been deduced to be dimers and/or oligomers of hematoporphyrin and its dehydrated derivatives.¹⁾ Synthetic studies of many different types of porphyrin dimers and

oligomers, therefore, have been carried out to clarify the tumor-localizing components of HPD and to obtain new effective photosensitizers for PDT.^{2,3)} HPD has, however, only weak absorption (ϵ 3000) in the Q band region (630 nm), although red light (> 600 nm) is effective in PDT because of its better tissue penetration.⁴⁾ A great number of dyes such as phthalocyanine, purpurin, chlorin e6, bacteriochlorin and benzoporphyrin, which have a strong absorption (ϵ 10^4 – 10^5) at longer wavelengths (> 650 nm), have been reported as potential photosensitizers during the past several years.⁵⁾

We chose chlorin e6 as a lead compound to generate new agents for PDT because of its abundance in green plants. In our previous paper, we reported the synthesis, physicochemical properties and photocytotoxicity of new δ -substituted chlorin e6 derivatives, and demonstrated that the triplet lifetime of these derivatives played an important role in their photocytotoxicity.⁶⁾ Our continuous efforts to obtain new photosensitizers for PDT have recently led to the syntheses of a new chlorin e6 dimer and trimer bridged with amide bonds on the analogy of HPD.

Although syntheses of ester- or ether-linked porphyrin dimers are more popular,²⁾ these dimers consist of several stereoisomers and are susceptible to bond cleavage. We therefore planned the syntheses of amide-linked chlorin e6 dimers without stereoisomers. First, we introduced an amino group into chlorin e6.

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Since pheophorbides have been reported to react with amines to yield chlorin-6-carboxamides by the cleavage of ring V,⁷⁾ methyl pheophorbide **a** (**1a**) was treated with a large excess of 1,3-propanediamine in chloroform to give the desired amine (**2a**) in a 72% yield.

A symmetrical dimer (**3b**) [FAB-MS m/z 1287 (MH)⁺; ¹H-NMR δ (CDCl₃) ppm: 7.10 (2H, m), 4.02 (2H, m), 3.82 (2H, m), 2.25 (2H, m) for the 1,3-propanediamine part] was obtained by the aminolysis of **1a** with **2b** in dry pyridine in a 48% yield; condensation of the amine (**2b**) with chlorin e6 6, γ -dimethylester (**4**) using water-soluble carbodiimide (WSC) in chloroform gave an unsymmetrical dimer (**5**) [FAB-MS m/z 1287 (MH)⁺; ¹H NMR δ (CDCl₃) ppm: 6.71 (1H, t, $J=5.9$ Hz), 5.82 (1H, t, $J=6.2$ Hz), 3.48 (1H, m), 3.27 (1H, m), 3.17 (1H, m), 3.12 (1H, m), 1.58 (2H, m) for the 1,3-propanediamine part] in a 54% yield. The assignments of these protons were established by ¹H-¹H COSY. This method is applicable not only to the syntheses of various chlorin e6 dimers like **3a** and **3c**, but also to those of chlorin e6 oligomers. Condensation of the amine (**2b**) with pheophorbide **a** (**1b**) using WSC gave a corresponding dimer (**6**) [FAB-

MS m/z 1255 (MH)⁺] in a 67% yield. Treatment of **6** with 1,3-propanediamine gave a corresponding amine (**7**) [FAB-MS m/z 1330 (MH)⁺] in a 70% yield, whose condensation with **4** using WSC gave a trimer (**8**) [FAB-MS m/z 1935 (MH)⁺] in a 20% yield. Repetition of these procedures by using pheophorbide **a** (**1a**) and diamines would theoretically give many different chlorin e6 oligomers with bridging at positions 6 and 7.

Recent studies have indicated that modification of the substituents at positions 2 and 4 of hematoporphyrin dimers and oligomers caused remarkable change in the *in vivo* photodynamic activity.²⁾ The above-mentioned method is also useful to synthesize chlorin e6 dimers modified at position 2. Treatment of 1,3-propanediamine with 2-(2-acetoxyethyl)-2-devinyl-pheophorbide **a** (**1c**), which was derived from methyl pheophorbide **a** (**1a**) by thallation, acid treatment, SBH reduction and acetylation,⁸⁾ gave a corresponding amine (**2d**), which resulted in a dimer (**3d**) by aminolysis with **1c**. The structures of **1a**, **1b**, **1c**, **3a**, **3b**, **3c** and **5** were confirmed by UV, ¹H-NMR, FAB-MS and elemental analysis. For **2a**, **2b**, **2c**, **2d**, **3d**, **4**, **6**, **7** and **8**, structural confirmation was

achieved by UV, ^1H NMR and FAB-MS. FAB-MS was measured by using 3-nitrobenzylalcohol as a matrix.

The *in vitro* and *in vivo* photodynamic activities of these chlorin e6 dimers and oligomers are under investigation.

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