PRELIMINARY STUDY ON PORPHYRIN DERIVATIVES AS TRANSFECTION REAGENTS FOR MAMMALIAN CELL

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Dedicated to:

My beloved mother and father

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ABSTRACT

Porphyrins are organic, aromatic compounds found in heme, cytochrome, cobalamin, chlorophyll and many other natural products with essential roles in biological processes that their cationic forms has been used as a groups of favorable non-viral vectors recently. Cationic porphyrins are self-chromogenic reagents with high capacity for modifications, great interaction with DNA and protection of DNA from nuclease during delivery of it into cell with low toxicity. In order to have high efficient gene transfection into cell while causing low toxicity, genetically manipulations of non-viral vector, cationic porphyrin, would be useful. In this study newly modified cationic porphyrins namely, 5-hexyl-10,15,20tris (*N*-methyl-4-pyridyl) porphyrin, 5-propyl-10,15,20tris (*N*-methyl-4-pyridyl) porphyrin, 5,10-dipropyl-15,20-bis (*N*-methyl-4-pyridyl) porphyrin, 5,10-dihexyl-15,20bis (*N*-methyl-4-pyridyl) porphyrin, and polyamidoamine (PAMAM) G_4-porphyrin conjugate were applied. Cytotoxicity of synthesize cationic porphyrins on Chinese Hamster Ovarian (CHO) cells, were evaluated by using MTT assay. Generally, all cationic derivatives are dose dependent, with low cytotoxicity at the ranges from 100 µM to 0.01µM. Four of cationic porphyrin were uptake by cell at high concentration while none were observed on conjugate one. Using different concentration of cationic porphyrins and methods were tested on transfection of CHO cells by using the derived transfection reagent with X-tremeGENE HP DNA as positive control. However no transfection observed by all the porphyrin derivatives and the parameters tested except for positive control. Results of this study suggested that applying different protocol, and also trying other concentration of cationic porphyrins and DNA for forming a strong complex would increase the possibility of efficient gene transfection by using cationic porphyrins.

ABSTRAK

Porphyrin ialah sebatian aromatic organik yang boleh ditemui di dalam hem, sitokrom, kobalamin, klorofil dan pelbagai lagi produk semulajadi. Dengan ciri kation, ia mempunyai fungsi yang penting dalam proses biologi semulajadi dan boleh dimanipulasi sebagai vector bukan viral dalam proses transfeksi. Porphyrins kation ialah bahan kimia kromogenik kendiri yang mempunyai cirri-ciri seperti kapasiti yang tinggi untuk diubahsuai, interaksi yang baik dengan DNA dan dapat melindungi DNA daripada nuklease semasa pemindahannya kedalam sel dibawah toksisiti yang rendah terhadap sel. Untuk mendapatkan transfeksi gen yang cekap kedalam sel dengan toksisiti yang rendah serta vektor bukan virus untuk pengubahsuai genetic, kationik porphyrin mungkin berguna untuk tujuan ini. Dalam kajian ini, kationik porphyrins yang digunakan ialah 5-hexyl-10,15,20tris (*N*-methyl-4-pyridyl) porphyrin, 5-propyl-10,15,20tris (*N*-methyl-4-pyridyl) porphyrin, 5,10-dipropyl-15,20-bis (*N*-methyl-4-pyridyl) porphyrin, 5,10-dihexyl-15,20bis (*N*-methyl-4-pyridyl) porphyrin, dan konjugat polyamidoamine (PAMAM) G_4-porphyrin. Kajian MTT digunakan untuk menentukan sitotoksisiti kationik porphyrins keatas Sel ovari hamster China (CHO). Secara umunya, kesemua terbitan kationik berkadar terus dengan dose toksisiti yang rendah dalam julat 100 µM to 0.01µM. Empat daripada kationik porphyrin telah diambil oleh sel pada kepekatan yang tinggi dan tidak pada sebatian konjugat. Dengan menggunakan kationik porphyrin pada kepekatan yang berbeza dan kaedah tranfeksi keatas sel CHO dan X-tremeGENE HP DNA telah digunakan sebagai kawalan positif. Walau bagaimanapun tiada transfeksi didapati oleh semua terbitan porphyrin dan parameter yang diuji kecuali kawalan positif. Keputusan kajian ini mencadangkan bahawa menggunakan protokol yang berbeza, dan juga julat kepekatan porphyrins kationik dan DNA yang lain untuk membentuk sebuah kompleks yang kukuh akan meningkatkan kemungkinan penggunaan porphyrins kationik sebagai transfeksi gen yang cekap.