



Preparation and evaluation of methylated N-(4-N,N dimethyl aminobenzyl) O- carboxymethyl chitosan as a non viral vector for gene delivery : *in vitro* studies

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Abstract: Chitosan is a biodegradable biopolymer emerging as a promising nonviral vector for gene delivery and due to its low toxicity and biocompatibility, chitosan received increasing attention as a versatile excipient. To improve water solubility and gene transfection efficiency of chitosan, we synthesized methylated N-(4-N,N dimethyl aminobenzyl) O- carboxymethyl derivative in this study as a novel gene delivery vector. After synthesizing the new derivative in three steps and determining the degree of substitution via H-NMR, nanoparticles of polymer and DNA plasmid were formed by ionic gelation method at different N/P ratios and then DNA retardation and DNase I tests were done. Finally zeta potential and size of selected N/P ratios were measured. In DNA retardation test the new derivative could retard pDNA migration at N/P ratios > 10 and in DNase I test pDNA was protected from enzyme degradation at N/P ratios of 10, 15, 20 and 25. According to the particle size and zeta potential measurements of polyplexes at these N/P ratios, the sizes are 234, 300, 217, 1182 nm and zeta potentials are 12.6, 9.20, 38.9 and 23 mV, respectively. Through these analyses the N/P ratios of 10 and 20 are selected and will be tested for ability of gene transfection and cytotoxicity in MCF-7 cell lines. In this study we showed that methylated N-(4-N,N dimethyl aminobenzyl) O- carboxymethyl chitosan as a novel vector is more soluble in water and can interact with pDNA and protect it from enzymatic degradation at lower N/P ratios and thus it would be a promising nonviral vector for *in vivo* gene delivery studies.

Keyword: chitosan, gene delivery, MCF-7 cell line