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MECHANISMS OF CELLULAR UPTAKE WITH CHITOSAN/DNA COMPLEX IN HEPATOMA CELL LINE

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Abstract

Chitosan (CS) has a high potential for gene delivery into mammalian cells. However, its uptake mechanism is not well clarified. We investigated the effects of inhibitors of clathrin-mediated endocytosis (chlorpromazine), caveolae-mediated endocytosis (genistein), macropinocytosis (LY 29004 and wortmannin), microtubuli polymerization (nocodazole) and of membrane cholesterol recycle (methyl-\(\beta\)-cyclodextrin) on the transfection efficiency with CS/pEGFP complexes and on the internalization of CS/rhodamine-labeled pEGFP complexes by hepatoma cell line (Huh 7 cells). The transfection was blocked by nocodazole, genistein, and methyl-\(\beta\)-cyclodextrin, respectively. CS/DNA complexes internalization was clearly inhibited by genistein. We conclude that the complexes uptake predominantly by caveolin-mediated pathways. In addition, fluorescence colocalization studies with acidotropic probes, LysoSensor\textsuperscript{TM} dye, illustrated that CS/DNA complexes are targeted to lysosomes for the degradation after internalization.