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Intracellular Trafficking and Drug Release from Fluorescently-Labeled Chitosan Nanoparticle Systems for Development of Innovative Drug Delivery Systems

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Abstract. The increased bioavailability of essential biomolecules such as drugs, DNA and peptides is pre-requisite for efficient intracellular efficacy on drug delivery systems. Nanotechnological-based approaches for drug delivery applications potentially promotes a better distribution of energy *in vivo*, increasing the intracellular uptake of biomolecules for enhanced therapeutic uptake. Realising the ubiquitous utilization of nanoparticles in an increasing myriad of research fields, investigations into nanoparticle uptake, cargo release, as well as nanoparticle carrier persistence are pertinent towards their consequent optimization and development. We describe in this work, the elucidation of nanoparticle uptake and sustained release of its encapsulated cargo in colon cancer cells to model a nanoparticle-mediated drug delivery system. Chitosan nanoparticles were synthesized through ionic gelation routes and characterized by means of light scattering, electron microscopy, and infrared spectroscopic analysis. The nanoparticles were encapsulated with a fluorescently-modified amino acid for *in vitro* tracking, and its intracellular release was quantitated in a time-dependent study using flow cytometry and fluorescent microscopy. Cytotoxic analysis was subsequently performed to evaluate any inherent efficacy of the nanoparticle for use as a candidate delivery system. Findings arising from our analyses showed that intracellular uptake of nanoparticles occurred within 30 mins of cell treatment; and continually took place up to 48 hours post treatment. Interestingly, release of cargo only occurred 6 hours post treatment and a controlled release system was exhibited up to 48 hours without extracellular leakage. MTT assay showed very low toxicity of the 60-180nm size particles; demonstrating a potential of the chitosan nanoparticle system for use as a systemic, slow release system for drug delivery. Conclusions derived from this study is hoped to provide sufficient data towards more critical developments of nanoparticle delivery systems for targeted and enhanced drug delivery parameters, most clinically relevant in the pharmaceutical and medical fields.

Keywords: nanoparticles, nanobiotechnology, chitosan, drug delivery systems, nanoparticle tracking