In vitro cellular localization and effcient accumulation of fuorescently tagged biomaterials from monodispersed chitosan nanoparticles for elucidation of controlled release pathways for drug delivery systems

ABSTRACT

Background Inefficient cellular delivery and poor intracellular accumulation are major drawbacks towards achieving favorable therapeutic responses from many therapeutic drugs and biomolecules. To tackle this issue, nanoparticle-mediated delivery vectors have been aptly explored as a promising delivery strategy capable of enhancing the cellular localization of biomolecules and improve their therapeutic efficacies. However, the dynamics of intracellular biomolecule release and accumulation from such nanoparticle systems has currently remained scarcely studied. Objectives The objective of this study was to utilize a chitosan-based nanoparticle system as the delivery carrier for glutamic acid, a model for encapsulated biomolecules to visualize the in vitro release and accumulation of the encapsulated glutamic acid from chitosan nanoparticle (CNP) systems. Methods CNP was synthesized via ionic gelation routes utilizing tripolyphosphate (TPP) as a cross-linker. In order to track glutamic acid release, the glutamic acid was fluorescently-labeled with fluorescein isothiocyanate prior encapsulation into CNP. Results Light Scattering data concluded the successful formation of small-sized and mono-dispersed CNP at a specific volume ratio of chitosan to TPP. Encapsulation of glutamic acid as a model cargo into CNP led to an increase in particle size to >100 nm. The synthesized CNP exhibited spherical shape under Electron Microscopy. The formation of CNP was reflected by the reduction in free amine groups of chitosan following ionic crosslinking reactions. The encapsulation of glutamic acid was further confirmed by Fourier Transform Infrared (FTIR) analysis. Cell viability assay showed 70% cell viability at the maximum concentration of 0.5 mg/mL CS and 0.7 mg/mL TPP used, indicating the low inherent toxicity property of this system. In vitro release study using fluorescently-tagged glutamic acids demonstrated the release and accumulation of the encapsulated glutamic acids at 6 hours post treatment. A significant accumulation was observed at 24 hours and 48 hours later. Flow cytometry data demonstrated a gradual increase in intracellular fluorescence signal from 30 minutes to 48 hours post treatment with fluorescently-labeled glutamic acids encapsulated CNP. Conclusion These results therefore suggested the potential of CNP system towards enhancing the intracellular delivery and release of the encapsulated glutamic acids. This CNP system thus may serves as a potential candidate vector capable to improve the therapeutic efficacy for drugs and biomolecules in medical as well as pharmaceutical applications through the enhanced intracellular release and accumulation of the encapsulated cargo.

Keyword : Drugs delivery; Nanotechnology; Chitosan; Glutamic acid; FITC