DEVELOPMENT OF MULTIFUNCTIONAL POROUS SILICA NANOPARTICLES FOR TARGETED AND STIMULI-RESPONSIVE GLIOBLASTOMA DRUG DELIVERY

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Background: Porous silica nanoparticles (PSiNPs) are among the most promising of drug delivery vehicles; made of chemically inert silica, their biocompatibility, large internal pore volumes and surface area, and ease of functionalisation, have long attracted attention in medical research. There is still, however a need for simple, scalable, and cost-effective methods to obtain multifunctional PSiNPs suitable for targeted cancer drug delivery. PSiNPs are most commonly synthesised through a modified Stöber method, a sol-gel process with cytotoxic surfactant CTAB acting as a template for the porous structure, although this method does not typically allow the synthesis of sub-100 nm particles required for optimum cell uptake. Protocols which allow access to the sub-100 nm range typically either lead to poor monodispersity, poor reproducibility, or require complex synthetic protocols. Furthermore, these particles then require post synthetic treatment to remove cytotoxic reagents and incorporate targeting and triggered drug release functionality rendering them unlikely candidates for clinical translation. This work presents a facile, reproducible, scalable and cost effective method for the synthesis of sub-100 nm PSiNPs, which incorporate cancer targeting and stimulated drug release without the need for retrospective functionalisation.

Methods: A novel synthetic pathway was developed where the particle size, uniformity and porous structure were templated by polyelectrolyte complexes formed from poly(acrylic acid) (PAA) and L-arginine (Arg). The addition of silica precursors lead to the formation of monodispersed porous silica nanoparticles functionalised with PAA and Arg. Drug loading capacities and release rates were determined with doxorubicin hydrochloride (Dox) and the efficacy of particle targeting and toxicity were tested *in vitro* using FITC-labeled particles in primary patient derived glioblastoma multiforme (GBM) and non-tumorigenic neural progenitor cells.

Results: The mixing of oppositely charged PAA and Arg led to the formation of monodispersed polyelectrolyte complexes ranging from 50-140 nm, which served as templates for the hydrolysis and condensation of the added silanes. The PSiNPs were readily tuneable from 40-200 nm while maintaining a narrow size distribution (PDI < 0.2), and with disordered pores ranging from 1-2.5 nm in diameter. The presence of PAA within the pores enabled high Dox loading, with a 22% w/w loading achieved, and provided a 4-fold increase in drug release over 48 hours under weakly acidic conditions (pH 5), representative of the endosomal and tumour microenvironments. The use of Arg in the polyelectrolyte complexes not only catalysed the basic hydrolysis and condensation of silane species but also conferred cancer targeting properties towards the cationic amino acid transporters which are overexpressed in GBM cells. The surface presentation of Arg gave significantly increased intracellular accumulation of PSiNPs in GBM1 and GBM20 cells compared to unfunctionalised PSiNP and to a non-tumorigenic control cell line ($P < 0.05$). The enhanced cellular accumulation effectively translated to a lower IC50 value of 2.9 μM compared to 8.1 μM of the non-targeted PSiNPs.

Conclusions: A new pathway for the synthesis of monodispersed, sub-100 nm PSiNPs with intrinsic functionality for stimuli-responsive drug delivery and tumour specific targeting was developed. This method obviates the need for cytotoxic reagents or post-synthetic functionalization associated with the conventional synthesis of PSiNPs. The simplicity and efficacy of this method presents a feasible candidate for progression into clinical trials and point-of-care medicine.