NOVEL CHITOSAN POLYMERIC MICELLES AS A DELIVERY VEHICLE OF HYDROPHOBIC ANTICANCER DRUGS

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Background: The synthesis of new chitosan derivatives with amphiphilic properties allows the production of polymeric micelles, which have the ability to encapsulate hydrophobic drugs as camptothecin (CPT). Once encapsulated, the drug is protected from the body fluids, its aqueous solubility is increased as well as its intestinal permeability and therapeutic activity.

Methods: Chitosan was chemically modified with O-methyl-O′-succinylpolyethylene glycol and with oleic acid by a carbodiimide reaction to produce micelles by self-assembly. Characterization of the copolymer included ^{1}H NMR, FTIR, GPC, DSC/TGA and XRD. Size, zeta potential and morphology were determined by DLS and TEM, respectively. The CPT association efficiency was determined by HPLC as well as its *in vitro* release and lactone ring protection from hydrolysis, both performed in simulated gastrointestinal fluids. Cytotoxic studies were performed against Caco-2 and HT29-MTX intestinal cell lines. CPT intestinal permeability was tested in three different *in vitro* cell models and biodistribution and pharmacokinetic studies were performed by gavage in male Balb/c nude mice colorectal cancer induced.

Results: The success of the synthesis and the purity of the new copolymer were demonstrated by ^{1}H NMR, FTIR and GPC and, after the grafting, the copolymer showed an increase on its thermal stability and crystallinity. The CMC revealed a good stability of the system after dilution and DLS revealed an average size of 140 nm, a positive superficial charge and CPT association efficiency of 78%. TEM analysis demonstrated a round and smooth shape for both empty and CPT-loaded micelles. The CPT *in vitro* intestinal release showed a low release in gastric media and a controlled release in intestinal fluids, suggesting a pH-dependent behaviour. Also, these micelles were able to protect CPT from hydrolysis up to 75% of its initial lactone form, exhibiting a good system stability. Regarding the safety profile, copolymer did not present a cytotoxic effect against colorectal cancer cell lines in concentrations equal or bellow 10 mg/mL. More importantly, CPT improved significantly its *in vitro* intestinal permeability, as compared with free CPT. Moreover, CPT-loaded micelles showed 5% tumor accumulation, a distribution volume of 0.3 L, an elimination half-life of 7.1 h and a clearance rate of 0.02 L/h.

Conclusions: CPT-loaded chitosan micelles proved to be a potential vehicle of hydrophobic anticancer drugs, as CPT.