Nonionic Polymeric Micelles for Oral Gene Delivery In Vivo

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Abstract

The main aim of this study was to investigate the feasibility of using nonionic polymeric micelles of poly(ethylene oxide)-poly(propylene oxide)-poly(ethylene oxide) (PEO-PPO-PEO) as a carrier for oral DNA delivery in vivo. The size and appearance of DNA/PEO-PPO-PEO polymeric micelles were examined, respectively, by dynamic light scattering and atomic force microscopy, and their ζ potential was measured. Expression of the delivered lacZ gene in various tissues of nude mice was assessed qualitatively by 5-bromo-4-chloro-3-indolyl-β-D-galactopyranoside staining of sections and quantitatively by measuring enzyme activity in tissue extracts, using the substrate of β-galactosidase, chlorophenol red-β-D-galactopyranoside. In addition, the types of cells expressing the lacZ gene in the duodenum were identified by histological analysis. DNA/PEO-PPO-PEO polymeric micelles are a single population of rounded micelles with a mean diameter of 170 nm and a ζ potential of -4.3 mV. Duodenal penetration of DNA/PEO-PPO-PEO polymeric micelles was evaluated in vitro by calculating the apparent permeability coefficient. The results showed a dose-independent penetration rate of (5.75 ± 0.37) x 10-5 cm/sec at low DNA concentrations (0.026-0.26 μg/μl), but a decrease to (2.89 ± 0.37) x 10-5 cm/sec at a concentration of 1.3 μg/μl. Furthermore, when 10 mM RGD peptide or 10 mM EDTA was administered before and concurrent with the administration of DNA/PEO-PPO-PEO polymeric micelles, transport was inhibited ([0.95 ± 0.57] x 10-5 cm/sec) by blocking endocytosis or enhanced ([29.8 ± 5.7] x 10-5 cm/sec) by opening tight junctions, respectively. After oral administration of six doses at 8-hr intervals, the highest expression of transferred gene lacZ was seen 48 hr after administration of the first dose, with gene expression detected in the villi, crypts, and goblet cells of the duodenum and in the crypt cells of the stomach. Reporter gene activity was seen in the duodenum, stomach, and liver. Activity was also seen in the brain and testis when mice were administered 10 mM EDTA before and concurrent with DNA/PEO-PPO-PEO polymeric micelle administration. lacZ mRNA was detected in these five organs and in the blood by reverse transcription-polymerase chain reaction. Taken together, these results show efficient, stable gene transfer can be achieved in mice by oral delivery of

PEO-PPO-PEO polymeric micelles.