Target Specific and Long-Acting Delivery of Biopharmaceuticals Using Hyaluronic Acid Derivatives

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Statement of Purpose: Hyaluronic acid (HA) is a biodegradable, biocompatible, non-immunogenic, non-toxic, and non-inflammatory linear polysaccharide. HA has been used for various medical applications such as arthritis treatment, ocular surgery, tissue augmentation, drug delivery and tissue engineering [1]. In this work, HA has been extensively investigated as novel drug carriers for target specific and long acting delivery of protein, peptide, and nucleotide therapeutics [2].

Methods:

Bio-imaging of HA derivatives using quantum dots (QDots): HA-QDot conjugates were synthesized and exploited for *in vitro* and *in vivo* bio-imaging of HA derivatives for drug delivery applications.

Target-specific intracellular delivery of siRNA: Poly(ethyleneimine) [PEI] was conjugated with HA for target specific intracellular delivery of siRNA by HA receptor mediated endocytosis.

Long acting conjugation of peptide with HA: HA was conjugated with anti-Flt1 peptide and formyl peptide receptor like 1 (FPRL1) receptor specific peptide for the development of peptide therapeutics with a feasible efficacy and pharmacokinetic characteristics.

Sustained release formulation of protein drugs: In order to prolong the release period of protein drugs, HA hydrogels encapsulating protein drugs with minimized protein denaturation were successfully developed using Michael addition chemistry.

Results: Real-time bio-imaging of HA derivatives was carried out using QDots. HA-QDot conjugates with 35 mol% HA modification maintaining enough binding sites for HA receptors were mainly accumulated in the liver, while those with 68 mol% HA modification losing much of HA characteristics were evenly distributed to the tissues in the body (Figure 1). The results are well matched with the fact that HA receptors are abundantly present in the liver with a high specificity to HA molecules. Accordingly, slightly modified HA derivatives were used for target-specific intracellular delivery of nucleotide therapeutics and highly modified HA derivatives were used for long-acting conjugation of peptide and protein therapeutics (Figure 2).

HA-PEI conjugate was synthesized for LYVE-1 receptor mediated target specific intracellular delivery of siRNA. The outer-compartment of HA in siRNA/PEI-HA complex appeared to bind to the LYVE-1 on B16F1 cell membrane, which was up-taken to the cells by receptor mediated endocytosis. When anti-VEGF siRNA/PEI-HA complexes were administered by intra-tumoral injections, they efficiently suppressed the tumor growth resulting in reduced VEGF production. The results supported the potential applications of HA-PEI conjugate for target specific intracellular delivery of siRNA with enhanced serum stability and gene silencing efficiency.

On the other hand, HA derivatives were also used for long acting conjugation of peptide drugs. Water-soluble FPRL1 receptor specific peptide (CWRYMVm) drug was conjugated to aminoethyl methacrylated HA (HA-AEMA) by Michael addition. For water-insoluble peptide drugs, tetra-butyl ammonium salt of HA (HA-TBA) was prepared using an ion-exchange resin in water. Then, water-insoluble anti-Flt1 peptide (GGNQWFI) was conjugated to HA-TBA in DMSO by the amide bond formation between carboxyl groups of HA and amine groups of GGNQWFI. HA-Peptide conjugates showed feasible efficacy and pharmacokinetic characteristics.

HA has been also used as novel depot systems in the forms of physically and chemically crosslinked hydrogels for various protein drug delivery applications. Sustained release formulation of hGH using high molecular weight HA microparticles was successfully developed as a once-a-week injection formulation. In order to prolong the release period of protein drugs, selectively crosslinked HA hydrogels have been also developed as a novel depot system to encapsulate protein drugs (Figure 2).

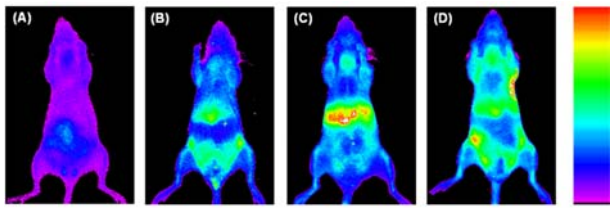

Figure 1. Fluorescent images of (A) a control, (B) QDots with an emission wavelength of 800 nm, and HA-QDot conjugates with (C) 35 mol% and (D) 68 mol% HA modifications.

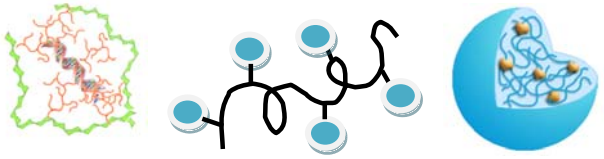

Figure 2. Target specific and long acting delivery applications of siRNA, peptide, and protein therapeutics using HA derivatives.

Conclusions: On the basis of real-time bio-imaging using QDots, HA derivatives have been successfully developed as novel drug carriers for target specific and long acting delivery of siRNA, peptide, and protein therapeutics. HA based drug delivery systems will be investigated further for clinical applications.

References:

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