Cartilage Lubrication through Peptide-mediated Hyaluronic Acid Binding and Retention

Anirudha Singh, Michael Corvelli, Heather Jacobs, Jennifer Elisseeff
Department of Biomedical Engineering, Wilmer Eye Institute, Johns Hopkins University, Baltimore, MD-21231

Statement of Purpose: Insufficient lubrication can lead to increased friction and degeneration of the articular cartilage. Viscosupplementation, a current clinical therapy, employs injection of viscous, high molecular weight HA, a natural lubricant in the body, to the joints. While it can temporarily reduce joint pain, the therapeutic effect of improving joint lubrication is limited due to a short joint residence time of HA. To address this, we developed a clinically relevant one-step injection technology that retains HA in the joints for a prolonged period of time and enhances the lubrication properties of normal and arthritic cartilage tissues.

Methods: Collagen and Hyaluronic acid binding peptides (HABPep) were conjugated to a PEG spacer. For lubrication studies, normal and arthritic cartilage surfaces were applied with the HABpep and HA, and tested using a parallel-plate Rheometer (ARES). For *in vivo* HA retention studies, HABpep and a fluorescently labeled HA were injected together to the rat knee joints and monitored with an IVIS imaging system.

Results: The data from our experiments suggested that the HABpep-polymer could capture HA that is found in low concentrations in a diseased environment or lost through a physical or biological mechanism, and provided the stable anchor on the tissue surface that was necessary to dynamically bind and concentrate HA on the surface. In rat normal joints, HA was retained for at least 72 h with the HABpep compared to the control (< 24 h).

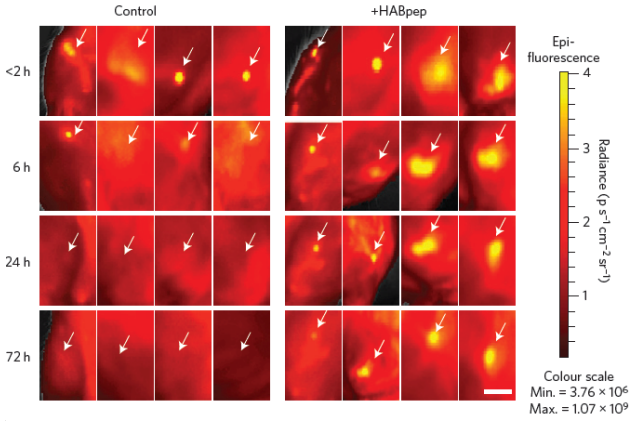

Figure 1: HA-rhodamine (white arrows) together with HABpep-PEG-Col IIBpep was injected into the healthy rat joints in a single step, and HA retention was monitored over time using an IVIS spectrum *in vivo* imager. Scale bar, 1 cm.

Normal cartilage treated with HA-binding coatings and pre-incubated in HA was able to replicate the low friction characteristics of native cartilage tested in an HA-rich environment. This pivotal result suggests that most of the lubrication effects of HA on the tissue can be replicated by surface-bound HA alone, without the need for large concentrations of HA in the local environment. Similar to the results from the normal cartilage surfaces, the OA cartilage samples treated with HABpep polymer coating that produces surface-bound HA produced static and

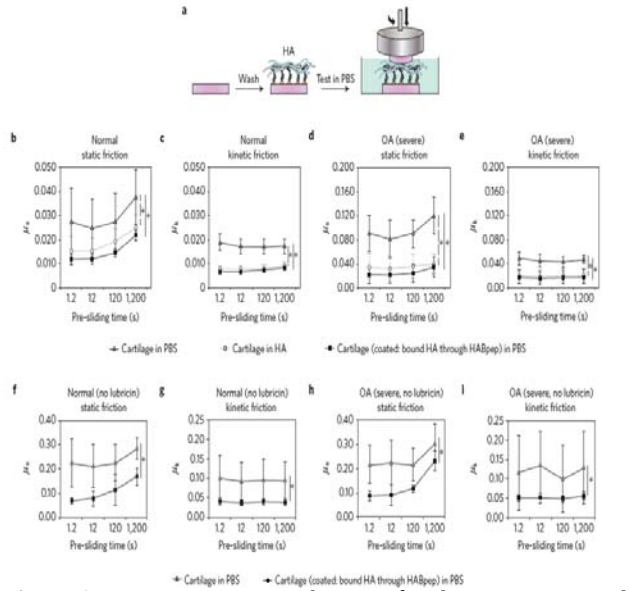

Figure 3: a, Representative schematic for the preparation and incubation of HABpep coated samples in test solution PBS. Lubrication properties of normal cartilage and severely damaged cartilage coated with the polymer-peptide system were tested in the presence of saline, and compared with uncoated surfaces in either saline or HA. Representative graphs of static friction and kinetic friction *vs.* pre-sliding time (s) for the normal cartilage sample (**b & c**) and severely damaged cartilage sample (**d & e**). (Statistical analyses: dashed lines represent cartilage samples (no HABpep modification) in PBS *vs.* HA bath and solid lines represent cartilage samples in PBS *vs.* cartilage samples coated with bound HA via HABpep in PBS.) Cartilage surface-bound HA via the HABpep-polymer coating system reduced friction values when lubricin is extracted from the tissue. Lubrication properties of normal cartilage and severely damaged cartilage (lubricin removed) coated with the polymer-peptide system was measured in PBS and compared to controls. Representative graphs of static friction and kinetic friction *vs.* pre-sliding time (s) for the normal cartilage sample (**f & g**) and severely damaged cartilage sample (**h & i**).

kinetic friction values nearly equal to those found with testing in an HA bath (Figs. 3). The practical implication is that even in a pathological environment, where low HA levels are present in the synovial fluid; the HA-binding coating can concentrate the limited HA available at the tissue surface to improve lubrication. Both normal and arthritic cartilage tissue benefited from the application of the HA binding technology with respect to lubrication and HA retention in the articular joint, suggesting that the technology is useful even in the presence of lubricin or could be synergistically applied with lubricin.

Conclusions: In summary, biomaterials-mediated strategies that locally bind and concentrate HA can provide physical and biological benefits to treat tissue-lubricating dysfunction and coat medical devices.

References: Singh A, *et al*. Nat. Mater. 2014;13:988-955.