Recombinant Human Hyaluronidase PH20 (rHuPH20) Facilitates Subcutaneous Dispersion and Systemic Absorption of Therapeutic Proteins in Non-Clinical Models

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ABSTRACT SUMMARY
Subcutaneous (SC) delivery of therapeutic proteins is an alternative to the traditional intravenous (IV) route of administration. It allows for less invasive delivery with key advantages, such as reduced incidence of systemic adverse effects and the ability to self-administer at home.1,2 However, current SC administration has disadvantages such as volume limitations, reduced flow rates, and potential adverse infusion site reactions, such as induration. Recombinant human hyaluronidase PH20 (rHuPH20) has been shown to facilitate SC delivery of viscous therapeutic proteins by improving local dispersion and systemic absorption, while reducing the incidence of induration and allowing larger volumes to be delivered at a single site.3

Immunoglobulin G (IgG) was used as a representative therapeutic protein and delivered subcutaneously into miniature pigs to evaluate the effects of rHuPH20 co-formulation. Endpoints included infusion pressures as measured by a reduction in interstitial fluid pressure (IFP), positron emission tomography (PET) imaging to measure volumetric dispersion and local clearance, and pharmacokinetic (PK) analysis to assess increases in systemic absorption.

The addition of 2000 U/mL of rHuPH20 significantly reduced IFP by 72% during a 20 mL SC infusion of a 15% IgG solution (p < 0.05). Additionally, PET imaging showed that rHuPH20 increased local volumetric dispersion of a 10% IgG solution by 22% within 0.5 hr and increased local clearance from the infusion site by 44% at 2 hr. Lastly, PK analysis indicated that the use of rHuPH20 reduced the Tmax of a 10% IgG solution by 35% and increased Cmax by 16%. The reduction in IFP, rapid dispersion and clearance, and increased rate of systemic absorption demonstrate that rHuPH20 facilitates SC administration of viscous therapeutic proteins.

INTRODUCTION
Conversion from intravenous to facilitated subcutaneous administration of therapeutic proteins can provide patients the option to self-administer at home, while shortening infusion times and reducing the incidence of systemic adverse effects. Currently, non-facilitated SC delivery is limited to small volumes that can be given at a single site and can be associated with potential local infusion reactions. However, SC infusions facilitated by recombinant human hyaluronidase PH20 (rHuPH20) have been shown to improve local SC dispersion, increase systemic absorption, and permit larger volumes to be delivered at a single site.

rHuPH20 is a recombinant, soluble form of the naturally occurring human hyaluronidase enzyme that transiently and locally degrades the substrate, hyaluronan (HA) in the extracellular matrix. The degradation of HA temporarily reduces the viscosity of the ‘gel-like’ phase of the SC extracellular matrix, allowing enhanced dispersion and systemic absorption of drugs delivered subcutaneously.

EXPERIMENTAL METHODS
Miniature pig models were used to assess infusion pressures, local fluid volumetric dispersion and clearance, and pharmacokinetic changes following a SC administration of IgG with and without rHuPH20. Studies were approved by institutional animal care and use committees, and animals were anesthetized with isoflurane prior to and during SC infusions in all experiments.

Assessment of Infusion/Interstitial Pressures: SC infusions were administered into the abdominal region of Yucatan miniature pigs. Infusions consisted of 20 mL of 15% human IgG ± rHuPH20 (2000 U/mL). Infusions were conducted using an infusion set with an 18 ga x ¾ inch winged infusion needle attached to a digital pressure transducer and a syringe filled with Control (IgG alone) or Test (with rHuPH20) article. A syringe pump was used to deliver the 20 mL volume at a flow rate of 2 mL/min while ‘real-time’ pressure measurements were recorded.

Assessment of Local Subcutaneous Volumetric Dispersion and Clearance by PET Imaging: Ibritumomab tiuxetan was radiolabeled with gallium-68 and co-mixed with 10% human IgG ± rHuPH20 (2000 U/mL). Control and Test articles (10 mL) were simultaneously injected (10 mL/min) into the abdominal region of miniature pigs. A PET imager was used to perform emission scans in dynamic mode followed by a transmission scan. The total, maximum, and average counts were analyzed within a fixed volume of interest (VOI) at each injection site.

Pharmacokinetic (PK) Analysis of Human IgG Absorption Following Subcutaneous Administration:
10 mL of 10% human IgG ± rHuPH20 (2000 U/mL) were administered SC at a flow rate of 4 mL/min into the abdominal region of miniature pigs. Plasma samples were collected at defined time intervals, and human IgG concentrations were measured via an ELISA. PK parameters were generated by analysis of concentration versus time data.

RESULTS AND DISCUSSION

Human IgG (10-15%) was used as a representative therapeutic protein to assess rHuPH20-facilitated SC infusions in pigs. Infusion volumes ranged from 10 to 20 mL per infusion site. Co-administration of rHuPH20 reduced infusion pressures, increased volumetric dispersion, and improved systemic absorption.

Assessment of Infusion/Interstitial Pressures:
Control infusions of 15% IgG alone produced an overall mean pressure of 83.1 mmHg throughout the course of a 20 mL infusion, while infusions of IgG with rHuPH20 resulted in a significantly reduced mean pressure of 23.3 mmHg (Fig. 1; p < 0.05).

Figure 1: Infusion/Interstitial Pressures

Assessment of Local Subcutaneous Volumetric Dispersion and Clearance by PET Imaging:
rHuPH20-facilitated infusions of 10% IgG resulted in a mean computed dispersion volume (1139 Voxels) at 25 min post-infusion, while infusions of IgG alone resulted in a significantly reduced mean dispersion volume (936 Voxels) at the same time point (p < 0.05). At 126 min post-infusion, IgG alone infusions had no significant change in local dispersion or clearance as seen by a mean volume (960 Voxels). In contrast, infusions with rHuPH20 resulted in significant local infusion site clearance with a mean volume (537 Voxels) at the same time point (Fig. 2; p < 0.05).

Figure 2: Local Dispersion and Clearance

**Pharmacokinetic (PK) Analysis of Human IgG Absorption Following Subcutaneous Administration:**

Plasma samples were analyzed at defined time intervals with the most significant changes observed within the first 18 hr. Infusions of 10% IgG alone resulted in a T<sub>max</sub> of 66 hr and a C<sub>max</sub> of 310 μg/mL. In contrast, rHuPH20-facilitated infusions reduced the T<sub>max</sub> to 43 hr, while increasing C<sub>max</sub> to 371 μg/mL (Fig. 3).

Figure 3: Human IgG Concentrations versus Time

**CONCLUSION**

Preclinical models were used to assess the effect of rHuPH20-facilitated subcutaneous infusions of a human IgG. rHuPH20 significantly reduced infusion pressures during a 20 mL SC infusion of 15% IgG. Additionally, rHuPH20 increased local volumetric dispersion and local clearance of 10% IgG as measured by PET imaging. Finally, rHuPH20 improved systemic absorption of 10% IgG by reducing T<sub>max</sub> and increasing C<sub>max</sub>.

Recombinant human hyaluronidase PH20 has been shown to facilitate subcutaneous administration of large volumes of therapeutic proteins at a single site.

**REFERENCES**


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