



Department of Health and Human Services
Public Health Service
Food and Drug Administration
Center for Biologics Evaluation and Research

BLA: 125402

Cross references: IND 13840
STN BL 125105 (GAMMAGARD LIQUID Immune Globulin Intravenous (Human), 10% Solution)
NDA 21-859 (Hylenex, (hyaluronidase human injection))

From: Evi Struble, Ph.D., Pharmacologist, CBER, DH, LPD

Through: Dorothy Scott, M.D., Laboratory Chief, CBER, DH, LPD

CC: Jennifer Reed, PhD
Mark Shields, RPM

Applicant: Baxter Healthcare Corporation

Product: Immune Globulin Infusion (Human), 10% with Recombinant Human Hyaluronidase; Proposed proprietary Name: HYQVIA

Subject: Preclinical Pharm-Tox Mid-cycle Review

Recommendations

There are no preclinical issues that would prevent this application from being approved.

Table of Content

<u>1. Background</u>	2
1.1 Indication	2
1.2 Dose	2
<u>2. Mode of Action as it is Presently Known</u>	2
2.1 Receptor/Binding	3
2.2 Species Cross Reactivity	3
2.3 Tissue Cross Reactivity	3
<u>3. The Principal Model(s) Used for Efficacy, <i>in vivo</i></u>	3
3.1 Primary Pharmacodynamics	3
3.2 Secondary Pharmacodynamics	3
<u>4. Toxicology</u>	4
4.1 Combination Product	4

4.2 rHuPH20	4
Margin of Safety	4
Conclusions, Toxicology Studies	5
5. PK Studies.....	6
Combination Product	6
RHuPH20 Alone	6
6. Distribution and Metabolism	7
7. References.....	7

1. Background

This BLA is for SC IG 10% co-packaged and administered in combination with recombinant human hyaluronidase PH20 (rHuPH20).

The immune globulin component of this product is identical to Gammagard Liquid 10% approved in 2005 under BLA 125105 for IV administration.

The recombinant human hyaluronidase PH20 (rHuPH20) component was also approved in 2005 under the trade name HYLENEX (hyaluronidase human injection). It is indicated as an adjuvant to increase the absorption and dispersion of other injected drugs; for subcutaneous fluid administration; and as an adjunct in subcutaneous urography for improving resorption of radiopaque agents. RHuPH20 is synthesized in Chinese hamster ovary (CHO) cells that have been transfected with a plasmid containing the DNA sequence encoding human hyaluronidase PH20. The approved preparation is made with what the submission refers to as “process (b)(4), which results in rHuPH20 concentrated at 160 U/mL.------(b)(4)-----

-----.

1.1 Indication

HyQvia is indicated for the treatment of patients with primary immunodeficiency (PI) associated with defects in humoral immunity.

1.2 Dose

The recommended dose of Immune Globulin Infusion (Human), 10% of HyQvia for patients with PI is 300 to 600 mg/kg body weight infused at 3 to 4 week intervals. However, in the clinical trial, higher doses were used, with a maximum dose of 1.8 mg/kg per 4 weeks (Table 14.3-28, Clinical Study 160603).

The amount of rHuPH20 co-packaged with IgG is 80 U/g IgG. Thus, for individuals taking the recommended dose of 0.6 g/kg IgG per month, the corresponding dose for rHuPH20 is 48 U/kg. Given the specific potency of the rHuPH20 preparation of ---(b)(4)---/mg the dose of rHuPH20 becomes --- (b)(4) ---.

For off-label uses, IgG up to 2 g/kg can be used, corresponding to a dose of rHuPH20 of 160 U/kg or 0.0013 mg/kg.

2. Mode of Action as it is Presently Known

Human PH20 hyaluronidase depolymerizes hyaluronan (HA) at neutral pH. Hyaluronan is a glycosaminoglycan polymer with MW 10^7 Da whose main function is to create a barrier to bulk fluid flow in the extracellular matrix (ECM).

HA rapidly turns over in the body. For example, 50% of the HA in the body is in the skin where it has a half life of less than 1.5 days (1). Most of the HA is degraded in the ECM to slightly smaller molecules of HA (MW 10^6 Da) resulting in increased mobility and migration to the

lymph nodes where they are further degraded and cleared. Up to 85% of HA is cleared in the lymphatics via the HA receptor for endocytosis, HARE (2). The rest 15% of HA enters the circulation through the thoracic duct, and it is cleared by the endothelial liver sinusoidal cells (LSC), and to a lesser extent, spleen (3, 4).

PH20 may play a role in water re-absorption in kidney (5), or may have other functions, but the mechanisms are not yet known.

2.1 Receptor/Binding

Hyaluronan (HA)

2.2 Species Cross Reactivity

PH20 is conserved across mammalian species. Based on a BLAST sequence alignment performed by this reviewer 90% homology or higher with mice, rat, dog, and monkey at the protein sequence level was noted.

2.3 Tissue Cross Reactivity

RHuPH20 binds to HA present in ECM such as in skin, cartilage, bone etc. Following subcutaneous injection rHuPH20 in animals there is no (or very little) exposure of the biologic in the blood.

3. The Principal Model(s) Used for Efficacy, *in vivo*

3.1 Primary Pharmacodynamics

Study No	Brief Description	Species
R03002	----(b)(4)----- assay	Mouse, ---(b)(4)-----

Permeation enhancement was tested by sc injection of *either* rHuPH20 ----(b)(4)----- Hyaluronidase reference standard (RS) *and* ---(b)(4)----- into (b)(4) mice. rHuPH20 temporarily increased -----(b)(4)----- dose-dependent fashion and showed equivalent activity as the RS.

3.2 Secondary Pharmacodynamics

Study No	Brief Description	Species
R03003	Dermal reconstitution	Mouse, ---(b)(4)-----
R04002	----(b)(4)--- assay after IV administration	Mouse, , ---(b)(4)-----
R04013	----- (b)(4)----- assay	Mouse, , ---(b)(4)-----
R08104	Dermal reconstitution	Mouse, ----(b)(4)-----
R08107	----(b)(4)----- assay	Mouse, ----(b)(4)-----

Study R03003 showed that the action of rHuPH20 was transient after intradermal injection; its effects were no seen at 24 h after injection in (b)(4) mice skin as measured by area of ----(b)(4)-----.

Study R04002 showed that IV administration of rHuPH20 in (b)(4) mice, increased ----(b)(4)----- (injected ID) area of ---(b)(4)--- in a dose-dependent manner.

Study R040013 showed that, in mice, rHuPH20 induced ----(b)(4)----- enhancement depends on the size of particles co-administered with it (ID). Significantly increased ---(b)(4)-- was only seen for particles up to (b)(4) nm in diameter.

Study R08107 (“*in vivo* potency”) showed mostly similar effects of the (b)(4) rHuPH20 products (made by ----(b)(4)-----) as measured by ----(b)(4)----- area.

Study R08104 showed that area of ---(b)(4)----- was significantly larger for rHuPH20 (made by - ----(b)(4)-----) than the vehicle control by (b)(4) hours or more suggesting that dermal reconstitution was complete at (b)(4) hrs (for rHuPH20 made with (b)(4) process) or ----- ---(b)(4)-----.

4. Toxicology

4.1 Combination Product

Local tolerance studies in rabbits and (b)(4) mice were performed for IGSC, 10% with and without rHuPH20 using SC route of administration.

In rabbits (study ---(b)(4)----- the local tolerance of IgG, 10% (500 mg/kg) with and without rHuPH20 and after repeated subcutaneous application, was evaluated. The IgG administration was associated with local site inflammation that increased in severity after repeated administration. It is likely that this reaction is due to immune reaction of rabbit’s immune cells with anti-galactose α 1, 3 galactose antibodies in human IgG. Mild reactions were found in the group where rHuPH20 and saline was administered. The ---(b)(4) of the preparation did not affect these changes (Report ---(b)(4)-----).

In (b)(4) mice (Study R09131) the local tolerance to three once/week subcutaneous doses of Gammagard liquid 10% with and without rHuPH20 was evaluated.

In this study, reactions associated with the administration of rhPH20 with or without Gammagard were not judged to be different from the saline control.

4.2 rHuPH20

Table 1. Tabulation of toxicology studies with rHuPH20 (modified from submission)

Type of Study	Species and Strain	Method of Administration	GLP	Study No.
Single-Dose -(b)(4)-	Rat, (b)(4)	IV	No	03-007/ R03005
Repeat-Dose Toxicity -(b)(4)- Days 1, 4, 8	Monkey, -(b)(4)-	Peribulbar, SC, once daily	No	---(b)(4)----- R05015
Repeat-Dose Toxicity -(b)(4)- Days 1, 8	Monkey, -(b)(4)-	Peribulbar, SC, once daily	Yes	---(b)(4)----- R05014
Repeat-Dose Toxicity -(b)(4)- 7 days	Monkey, ----(b)(4)-----	IV and SC, once daily	Yes	---(b)(4)----- R08056
Repeat-Dose Toxicity -(b)(4)- 6 weeks	Monkey, ----(b)(4)-----	Intravesical, once weekly	Yes	1005-1253/ R05108
Repeat-Dose Toxicity -(b)(4)- 39 weeks	Monkey, ----(b)(4)-----	SC, once weekly	Yes	258.01/ R09050
Embryo-fetal Development -(b)(4)- DG 6-15	Mouse, ----(b)(4)-----	SC, once daily	No	---(b)(4)----- R07046
Embryo-fetal Development -(b)(4)- DG 6-15	Mouse, ----(b)(4)-----	SC, once daily	Yes	---(b)(4)----- R08176
Pre/postnatal Development -(b)(4)- DG 6-DL 20 or DG 22	Mouse, ----(b)(4)-----	SC, once daily	Yes	---(b)(4)----- R09058
Local Tolerance -(b)(4)- Days 1, 3, 7	Rat, (b)(4)	IP, once daily	No	04-007/ R05049

Margin of Safety

Table 2. NOAEL Determined in Toxicity Studies with rHuPH20 (modified from submission; includes a higher human dose) and the calculated Margin of Safety

Species	NOAEL (mg/kg)	HED ¹ (mg/kg)	Study Numbers	Margin of Safety ²	
				Based on HED	Based on mg/kg dose
--(b)(4)-- monkey	peribulbar: 0.01 subcutan: 0.1	0.003 0.032	----(b)(4)----- R05015	2 25	8 77
--(b)(4)-- monkey	0.1	0.032	----(b)(4)----- R05014	25	77
----(b)(4)----- monkey	0.64	0.206	1005-1253/ R05108	158	492
Rat (IP)	0.0125	0.002	R05049	1.5	10
----(b)(4)----- monkey	5	1.613	----(b)(4)----- R08056	1,240	3,846
----(b)(4)----- monkey	2	0.645	----(b)(4)----- R09050	496	1,538
Mice	3	0.244	----(b)(4)----- R07046	188	2,308
Mice (developmental)	maternal: 18 fetal: 3	1.463 0.244	----(b)(4)----- R08176	1,125 188	13,846 2,308
Mice (developmental)	9	0.732	----(b)(4)----- R09058	563	6,923

¹ Human Equivalent Dose (calculated based on relative body surface area)

² Compared to the maximum intended human dose of 0.0013 mg/kg body weight (based on off-label dose of 2 g/kg IgG, the rHuPH20 amount in the package of 80 U/g IgG, and specific activity of HuPH20 ----(b)(4)-----).

Conclusions, Toxicology Studies

1. There was an increase in splenic weight in ----(b)(4)----- monkeys following 39 week repeated SC administration of rHuPh20 (report R09050). No dose relationship was seen and no microscopic observations were noted. A dose-related spleen enlargement is seen in female mice at the end of study R07046, following repeated administration. These reactions could be due to immune response following repeated administration of the human recombinant protein.
2. Local reactions were seen following combination product and to a much smaller extent, following rHuPH20.
 - a. rhPH20 SC administration with or without Gammagard were not judged to be different from the saline control in (b)(4) mice (Study R09131).
 - b. IgG administration was associated with local site inflammation that increased in severity after repeated administration in rabbits (Report ----(b)(4)-----, likely due to immune reaction of rabbit's immune cells with anti-galactose α 1, 3 galactose antibodies in human IgG. Mild reactions were found in the group where rHuPH20 and saline was administered. The ----(b)(4) of the preparation did not affect the local changes.
 - c. Minimal, dose related subcutaneous perivascular lymphoplasmacytic infiltration of plasma cells and lymphocytes were seen in the rHuPH20 injection site at the two highest dose groups in ----(b)(4)----- monkeys (0.2 mg/kg, or ~150X human

- dose and 2 mg/kg) in study ----(b)(4)----- . Recovery was seen after the 4-week recovery period.
3. Slight renal tubule dilation with the lumina filled with “hyaline casts” is seen in male rats following single IV administration of rHuPH20 at a dose ~65X human dose (R03005). Uterine (hydrometra) and kidney toxicity (endothelial changes) were seen in female rats after single doses (~9X clinical dose) administered IP (Report R05049). Due to the different route of administration, it is not clear what the relevance of these findings would be in the clinic.
 4. There are signs of hepatotoxicity in male ----(b)(4)----- monkeys (Report 08056) following repeated (7 days) SC administration of rHuPH20, namely hepatocytic vacuolation in 2/2 males in the SC group with or without ALT, AST and CK elevation. This sign is not seen after IV administration in the same study or following 39 weekly SC dosing in ---(b)(4)----- monkeys.
 5. Repeated administration of rHuPH20 results in neutralizing antibody formation in ----(b)(4)----- monkeys. In study ----(b)(4)----- antibody titers correlated to dose and generally increased over time. After the 4-week recovery period, hyaluronidase neutralizing activity in most animals dropped approximately 50% from peak levels.
 6. Reproductive and development studies demonstrated that at high doses, rHuPH20 was embryofetotoxic but did not have a teratogenic effect.
 - a. The developmental NOAEL for the fetuses is 3 mg/kg/day in mice (study 08176). Reductions in fetal weight and increases in the number of late resorptions occurred in the 9 and 18 mg/kg/day dosage group. Total litter losses were seen at SC doses of 30 mg/kg/day in mice (study ----(b)(4)----- , report R07046).
 - b. Maternal NOAEL of rHuPH20 is 18 mg/kg/day (study 08176 in mice).
 7. Effects of rHuPH20 on fertilization were not performed. It is not known if anti-PH20 neutralizing antibodies would have any effect in fertilization in men or women.
 - a. Endpoints of reproductive organ morphology and function in male and female ----(b)(4)----- monkeys were included in the 39 week subchronic study. The NOAEL was determined to be 2 mg/kg, the highest dose tested.
 - b. The NOAEL for reproduction in the dams (i.e. animals who are already pregnant) and for viability and growth in offspring (i.e. F1 animals that were exposed to rHuPH20 *in utero*), including sexual maturation, learning and memory and the ability to produce an F2 generation is 9 mg/kg/day (study R09058 in mice).

5. PK Studies

Combination Product

Pharmacokinetics for IgG, 10% with rHuPH20 were investigated in rabbits and dogs.

In rabbits (Report ----(b)(4)----- , IgG 10% administered SC (500 mg/kg) with or without rHuPH20 or saline had no influence on any of the PK parameters of IgG, 10% in rabbits.

In dogs (Report R06019 & R06025), IgG 10% (1,000 mg/kg) administered SC with or without rHuPH20 at different ratios (as admix or sequentially) showed that:

1. Co-formulation of IgG with 100 U/mL rHuPH20 significantly increased C_{max} (146%) and decreased T_{max} to (30%) as compared to SC administration of IgG alone.
2. Sequential administration or mixture with lower amounts of rHuPH20 did not affect PK parameters.

RHuPH20 Alone

PK studies were performed in mice, rats, ----(b)(4)----- monkeys.

In rats (Report R05107), single IV doses of rHuPH20 (86, 860 and 8,600 U/kg) resulted in detectable plasma hyaluronidase activity at 1 min post-injection only in the highest dose, falling below the level of detection ((b)(4) U/mL) at 5 min post-injection.

In mice (Report R09023), single IV doses of 48,000 U/kg rHuPH20 of (b)(4) and (b)(4) preparations resulted in half-lives less than 5 minutes and plasma activity of rHuPH20 below lower limit of quantitation by 30 minutes.

In ----(b)(4)----- monkeys (Report R07060) it was shown that after single-doses of rHuPH20 (b)(4)--, the half-life of hyaluronidase activity in plasma was ~5 minutes and 10.3 to 15.7 hours after IV and SC administration respectively. Estimated bioavailability after SC administration is 1.5-4%. At SC doses of 1 mg/kg, most of the plasma PH20 activity were below the assay's quantification limit.

Toxicokinetic profiles were evaluated in repeated dose studies in ----(b)(4)----- monkeys (reports R08056 and R09050) confirming low systemic exposure after SC administration. As shown in report R09050, plasma activity of rHuPH20 at SC doses of 0.02 and 0.2 mg/kg was below baseline levels. Only for the animals that received the highest dose (2.0 mg/kg/week) of rHuPH20 (b)(4)--, plasma hyaluronidase activity exceeded pre-dose levels. Due to antibody formation at all dose levels, the C_{max} and AUC of PH20 after peaking on day 85 for M and 183 for F, decrease at later time points. The exposures are consistently higher for F than M. T_{max} at all time-points is 6 hours or less with most group average plasma levels below quantitation limit at 12 hours.

6. Distribution and Metabolism

Limited studies were performed with rHuPH20 regarding absorption and distribution following subcutaneous administration in animals.

37,000 U/kg rHuPH20 administered in female -----(b)(4)---- mice (report R08127) resulted in injection site activity half-life between 13 to 20 minutes - much shorter than plasma half life in ----(b)(4)----- monkeys following SC administration (~10-16 hrs). Low rHuPH20 activity was observed in lymph tissue homogenates up to 24 hours post dose.

Metabolism studies were not performed with rHuPH20. Like other glycoproteins and depending on the glycosylation pattern, PH20 is likely to be internalized intracellularly via glycan-binding lectins such as mannose, and asyalo-glycoprotein receptors and undergo proteolysis and catabolism. Likely sites of clearance are the injection sites, the draining lymph nodes, endothelial cells of blood vessels and the liver.

7. References

1. Tammi, R., Saamanen, A. M., Maibach, H. I., & Tammi, M. (1991) *J Invest. Dermatol.* **97**, 126-130.
2. Harris, E. N., Weigel, J. A., & Weigel, P. H. (2004) *J Biol. Chem.* **279**, 36201-36209.
3. Weigel, P. H. & Yik, J. H. (2002) *Biochim. Biophys. Acta* **1572**, 341-363.
4. Laurent, T. C. & Fraser, J. R. (1992) *FASEB J* **6**, 2397-2404.
5. Grigorieva, A., Griffiths, G. S., Zhang, H., Laverty, G., Shao, M., Taylor, L., & Martin-DeLeon, P. A. (2007) *Kidney. Blood. Press. Res.* **30**, 145-155.