CLINICAL PHARMACOLOGY BLA REVIEW
Division of Hematology
Office of Blood Review & Research

STN 125591

Sponsor: CSL Behring

Product: Antihemophilic Factor (Recombinant), rVIII-SingleChain

Indication: To treat patients with hemophilia A (congenital Factor VIII deficiency)

Submission Date: May 29, 2015

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INTRODUCTION

Single-chain recombinant Factor VIII (rVIII-SingleChain) is produced in Chinese hamster ovary (CHO) cells. rVIII-SingleChain is a single polypeptide chain with a truncated B-domain that allows for a covalent bridge to link the Factor VIII heavy and light chains. As compared with full-length Factor VIII, the stabilized single-chain design results in increased binding affinity to VWF. VWF stabilizes Factor VIII and protects it from degradation.

rVIII-SingleChain is a recombinant protein that replaces the missing Coagulation Factor VIII needed for effective hemostasis, is purified by a controlled multi-step process including two virus reduction steps complementing each other in their mode of action. No human or animal derived

proteins are used in the purification or formulation processes. rVIII-SingleChain is a preservative-free, sterile, non-pyrogenic, lyophilized powder to be reconstituted with water for injection for intravenous injection.

The number of units of Factor VIII administered is expressed in IU, which is related to the current WHO standard for Factor VIII products. One IU of Factor VIII activity in plasma is equivalent to that quantity of Factor VIII in 1 mL of normal plasma. Factor VIII activity in plasma is expressed either as a percentage (relative to normal human plasma) or in IU (relative to an International Standard for Factor VIII in plasma).

The following is the clinical pharmacology studies reviewed in this submission.

CLINICAL PHARMACOLOGY LABELING COMMENTS

1.1 12.1 Mechanism of Action

<<TRADENAME>> is a recombinant protein that replaces the missing Coagulation Factor VIII needed for effective hemostasis. <<TRADENAME>> is a single polypeptide chain with a truncated B-domain that allows for a covalent bridge to link the Factor VIII heavy and light chains. Compared to full-length Factor VIII, the stabilized single-chain design results in increased binding affinity to VWF. VWF stabilizes Factor VIII and protects it from degradation. Activated <<TRADENAME>> has an amino acid sequence identical to endogenous FVIIIa.

1.2 12.2 Pharmacodynamics

Hemophilia A is an x-linked hereditary disorder of blood coagulation due to decreased levels of Factor VIII and results in bleeding into joints, muscles or internal organs, either spontaneously or as a result of accidental or surgical trauma. Replacement therapy increases the plasma levels of Factor VIII enabling a temporary correction of the factor deficiency and correction of the bleeding tendencies.

1.3 12.3 Pharmacokinetics

Adults (≥18 years)

The pharmacokinetics (PK) of <<TRADENAME>> were evaluated in 81 previously treated subjects following an intravenous injection of a single dose of 50 IU/kg. Table 5 summarizes the PK parameters estimated based on plasma Factor VIII activity by chromogenic assay. The PK profile obtained 3 to 6 months after the initial PK assessment **(first dose)** was comparable with the PK profile obtained after the first dose. The **estimation of** PK parameters (Table 5) were based on plasma Factor VIII activity measured by the chromogenic assay. after the first dose (initial PK assessment).

Table 5: Pharmacokinetic Parameters (Arithmetic Mean, CV%) Following a Single Injection of 50 IU/kg of <<TRADENAME>> - Chromogenic Assay

PK Parameters	<<TRADENAME>> 50 IU/kg (N=81)
IR (IU/dL)/(IU/kg)	2.00 (20.8)
C_{max} (IU/dL)	106 (18.1)
AUC_{0-inf} (IU*h/dL)	1960 (33.1)
$t_{1/2}$ (h)	14.2 (26.0)
MRT (h)	20.4 (25.8)
CL (mL/h/kg)	2.90 (34.4)
V_{ss} (mL/kg)	55.2 (20.8)

IR = incremental recovery recorded at 30 minutes after injection; AUC_{0-inf} = area under the Factor VIII activity time curve extrapolated to infinity; $t_{1/2}$ = half-life; MRT = mean residence time; CL

= body weight adjusted clearance; V_{ss} = body weight adjusted volume of distribution at steady-state.

Adolescents and Children (<18 years)
Pharmacokinetics parameters of <<TRADENAME>> were evaluated in 23 previously treated subjects, 10 adolescents (≥12 to <18 years) and 13 children (0 to <12 years) in open-label, multicenter studies following a 50 IU/kg intravenous injection of <<TRADENAME>>. Table 6 summarizes the PK parameters estimated based on plasma Factor VIII activity measured by chromogenic assay. The body weight adjusted clearance of <<TRADENAME>> in children 0 to <6, ≥6 to <12, and ≥12 to <18 years of age was higher by 2, 1.74, and 1.31-fold than adults, respectively. The half-life of <<TRADENAME>> was shorter in children <12 years of age than adults by 4 to 5 hours.

Table 6: Comparison of Pharmacokinetic Parameters by Age Category (Arithmetic Mean, CV%) Following a Single Injection of 50 IU/kg of <<TRADENAME>> - Chromogenic Assay

PK Parameters	0 to <6 years	≥6 to <12 years	≥12 to <18 years
	(N=6)	(N=7)	(N=10)
IR (IU/dL)/(IU/kg)	1.52 (13.3)	1.56 (25.3)	1.69 (24.8)
C_{max} (IU/dL)	77.1 (12.7)	77.9 (25.3)	89.7 (24.8)
AUC_{0-inf} (IU*h/dL)	900 (19.9)	1060 (25.9)	1540 (36.5)
$t_{1/2}$ (h)	8.71 (27.6)	9.95 (7.9)	14.3 (33.3)
MRT (h)	10.5 (29.1)	12.6 (9.7)	20.0 (32.2)
CL (mL/h/kg)	5.91 (26.7)	5.06 (31.1)	3.80 (46.9)
V_{ss} (mL/kg)	72.4 (12.0)	72.3 (28.5)	68.5 (29.9)

IR = incremental recovery recorded at 30 minutes after injection for subjects 12 to < 18 years and at 60 minutes after injection for subjects 1 to < 12 years; AUC = area under the Factor VIII activity time curve extrapolated to infinity; $t_{1/2}$ = half-life; MRT = mean residence time; CL = body weight adjusted clearance; V_{ss} = body weight adjusted volume of distribution at steady-state.

Recommendations

The design, results, and interpretation of pharmacokinetic studies are acceptable. The sponsor has accepted the clinical pharmacology labeling change as requested by the FDA. The clinical pharmacology reviewer recommends the approval of AFSTYLA for the treatment of patients with hemophilia A (congenital Factor VIII deficiency).

Study #1

Study Title: A Phase I/III open-label, multicenter, crossover safety, efficacy and pharmacokinetic study of Recombinant Coagulation Factor VIII (rFVIII) Compared to Recombinant Human Antihaemophilic Factor VIII (rFVIII; INN: octocog alfa) in subjects with Hemophilia A, and a repeat PK, safety and efficacy study (CSL627-1001).

This was an open-label, international, multicenter, cross-over study and consisted of three parts (Figure 1). The objectives of the study were as follows:

Primary:
• Characterize the rate of inhibitor formation
• Characterize the PK profile of rVIII-SingleChain
• Demonstrate efficacy in the prevention and treatment of bleeding events
• Demonstrate the efficacy of routine prophylaxis treatment over on-demand treatment
• Demonstrate the efficacy of rVIII-SingleChain in surgical prophylaxis

Secondary:
• Characterize the safety profile of rVIII-SingleChain
• Perform the PK comparison of rVIII-SingleChain to Advate

Part 1: Eligible subjects included males of at least 18 years of age (18-65), who had been diagnosed with severe hemophilia A with <1% FVIII:C levels. There were 27 subjects in the study. This part of the study included a single-sequence crossover PK comparison of Advate and r-VIII SingleChain. Subjects received a single intravenous (IV) dose of 50 IU/kg Advate followed by the same dose of rVIII-SingleChain after a 4-day wash-out period. Advate was dosed based on the actual potency and rVIII-SingleChain based on the chromogenic substrate assay. Blood samples for PK assessment were collected at pre-dose, 0.5, 1, 4, 8, 10, 24, 28, 32, 48 and 72 h.

Part 2: This part of the study assessed efficacy and safety of rVIII-SingleChain with continued dosing from Part 1. The first 5 subjects received on-demand treatment to confirm the hemostatic potential of rVIII-SingleChain, while the remaining subjects received either on-demand or prophylaxis treatment based on their preference and investigator discretion.

Part 3: Eligible subjects included males ≥ 12 to 65 years of age, who had been diagnosed with severe hemophilia A and had < 1% FVIII:C levels. This part of the study assessed the safety and efficacy of rVIII-SingleChain with continued dosing of new subjects, and included a repeat PK assessment for at least 13 subjects. All subjects enrolled from Japanese sites participated in the
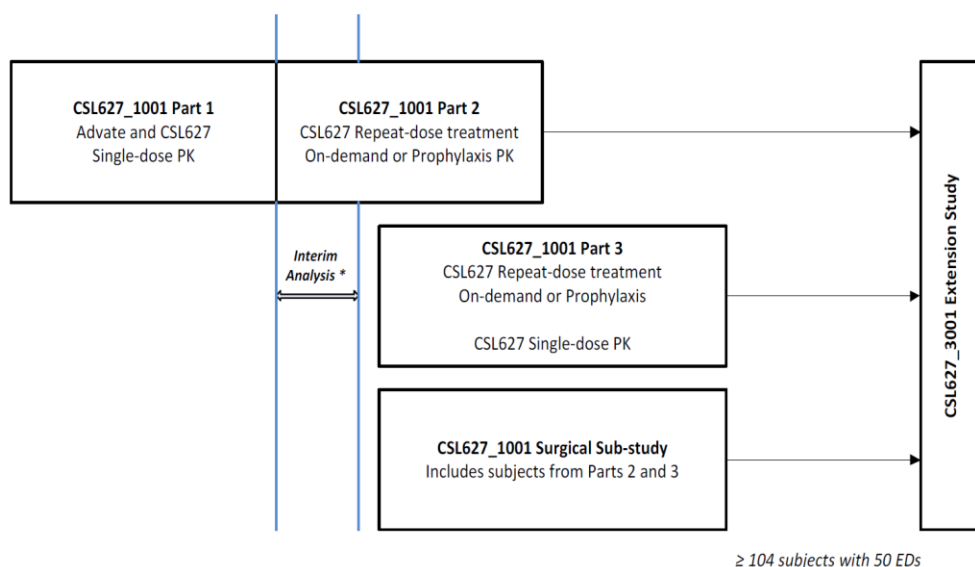
PK analysis. After PK assessment, subjects then began on-demand or prophylaxis treatment for at least 50 Exposure Days (EDs).

After enrollment commenced in Part 3, a surgical sub-study was conducted with a minimum of 5 subjects from either Parts 2 or 3. In this sub-study, dosing regimens with rVIII-SingleChain were individualized based on the type of surgery and clinical status of the subject.

Approximately 30 subjects were to be enrolled into Part 1, to ensure 26 evaluable subjects for the PK comparison. Approximately 100 additional subjects were be enrolled in Part 3, combined with the 26 to 30 subjects from Parts 1 and 2, to ensure that there are at least 104 evaluable subjects to characterize the inhibitor rate. There were 13 subjects in the PK analysis for surgical population. In part 3, formulations of two different strengths (250 and 3000 IU) were also evaluated (PK based).

PK analysis for rVIII-SingleChain was completed for 64 subjects enrolled and repeated between months 3 and 6. In part 3, blood samples for PK assessment were collected at pre-dose, 10-15 min, 0.5, 1, 3, 6, 9, 24, 32, 48, 72 and 96 h.

Figure 1: Study Design

≥ 104 subjects with 50 EDs

FVIII activity in human plasma was determined using 2 assays, chromogenic substrate (Chs) and one-stage (OS) assays, both of which use the (b) (4) A discrepancy between factor VIII activity measurements exist based on the assay method used. rVIII-SingleChain potency assignment was determined using the chromogenic substrate assay method and was the primary method of analysis for FVIII:C and PK evaluations. Non-compartmental analysis was used to estimate PK parameters.

PK parameters were calculated with and without correction for any pre-dose FVIII levels. For pre-dose correction baseline or pre-dose value before the first dose for each product was subtracted from all the post-injection measurements both after the first dose and repeat dose at the steady-state.

PK data from Part 1 of the study (PK parameters calculated from chromogenic substrate assay only) were analyzed to assess the comparative bioavailability of rVIII-SingleChain and Advate after dose-adjustment. The plasma FVIII activity measures (C_{max}, $AUC_{0\text{-last}}$ and $AUC_{0\text{-}\infty}$) were log-transformed and a 90% confidence interval (CI) for the ratio of the averages (population geometric means) of the FVIII activity measures for rVIII-SingleChain and Advate was applied. In order to establish comparative bioavailability, the upper and lower 90% CI must fall within the pre-established limits of 80-125%.

PK data from Part 3 of the study (PK parameters calculated from chromogenic substrate assay only) were also analyzed to assess comparative bioavailability between the low and high strength rVIII-SingleChain formulations. Same statistical approach was taken as described previously to demonstrate comparative bioavailability.

Table 1 is the demographic summary for PK study. The PK parameters of rVIII-SingleChain are shown in Tables 2-8. Concentration-time profiles of rVIII-SingleChain are shown in Figures 1-2. The results of the study can be summarized as follows:

- The PK of rVIII-SingleChain was characterized in subjects with severe hemophilia A. After a single IV dose of 50 IU/kg, the mean clearance and half-life of rVIII-Single Chain by chromogenic assay (Chs) were 2.55 ± 0.74 mL/hour per kg and 14.7 ± 3.7 hours, respectively. The mean clearance and half-life of rVIII-SingleChain by one stage assay (OS) were 4.13 ± 1.31 mL/hour per kg and 15.5 ± 4.9 hours, respectively. Although, the half-life of rVIII-SingleChain was not different between Chs and OS methods but the clearance was 1.6-fold higher by OS method than the Chs method. C_{max}, $AUC_{(0\text{-infinity})}$, and IR values by OS assay were 49%, 63%, and 48% of the values than those determined by the ChS assay (Table 2).

- The 90% confidence interval (90% CI) for C_{max} ranged from 90.5 to 104.6, indicating that C_{max} met the bioequivalence criteria. The 90% CI interval for $AUC_{(0\text{-infinity})}$ failed to meet the bioequivalence criteria (range = 117.8-159.9).

- The PK of single and repeat dose (3 to 6 months after the initial PK) of rVIII-Single Chain was comparable for all subjects (Table 3). However, in subjects ≥12 to <18 years of age, the clearance of rVIII-SingleChain was lower by 34% by OS and ChS analytical methods (Table 4). This difference between single and multiple dose may be an artifact and of little practical value. There were 10 and 5 subjects in the single and multiple dose study, respectively. There was also a high variability following a single dose (approximately CV = 47%) whereas multiple dosing had much lower variability (CV = 11-20%). This difference in variability may be due to the difference in sample size between single and multiple dosing.

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- Although the half-life of rVIII-SingleChain in subjects ≥12 to <18 years of age was comparable to those in subjects ≥ 18 years, clearance (based on per kg body weight) was 31% and 24 % higher in subjects ≥12 to <18 years of age than subjects ≥ 18 years of age by chromogenic assay and one stage assay, respectively (Tables 4 and 5).
- The PK parameters of of rVIII-SingleChain were comparable between non-Japanese and Japanese subjects (Tables 6 and 7).
- The FVIII PK was comparable between subjects dosed with of 250 IU strength and those dosed with 3000 IU strength rVIII-SingleChain formulation (Table 8).

Table 1: Demographics of pharmacokinetic population

	Part 1 (N=27)	Part 3 (N=64)
Age (years)		
Mean (SD)	35.4 (10.83)	28.3 (11.41)
Median	32.0	26.0
Min, Max	19, 60	12, 58
Age group (n [%])		
≥ 12 to < 18 years	0	10 (15.6)
≥ 18 to ≤ 65 years	27 (100.0)	54 (84.4)
Weight (kg)		
Mean (SD)	77.9 (11.46)	69.7 (19.82)
Median	76.6	65.2
Min, Max	59, 100	37, 112
BMI (kg/m^2)		
Mean (SD)	24.7 (3.19)	23.0 (5.13)
Median	25.3	21.1
Min, Max	19, 30	15, 37
BMI category (n [%])		
< 30 kg/m^2	27 (100.0)	57 (89.1)
≥ 30 kg/m^2	0	7 (10.9)
Race (n [%])		
Asian	0	23 (35.9)
Black or African American	2 (7.4)	11 (17.2)
White	25 (92.6)	29 (45.3)
Other	0	1 (1.6)
Ethnicity (n [%])		
Hispanic or Latino	5 (18.5)	2 (3.1)
Not Hispanic or Latino	22 (81.5)	62 (96.9)
Geographical region (n [%])		
United States	9 (33.3)	5 (7.8)
Japan	0	10 (15.6)
Europe	18 (66.7)	21 (32.8)
Rest of the world	0	28 (43.8)

BMI, body mass index; SD, standard deviation.

Notes:

[1] BMI is calculated as follows: BMI at Screening = Weight at Screening (kg) / (Height at Screening [m])^2.

[2] Percentages are based on the number of subjects in the respective group.

Conclusions: The half-life of rVIII-SingleChain by both analytical methods was comparable with Advate. The clearance of rVIII-SingleChain was 16% and 54% lower than advate by Chs and OS analytical methods, respectively. In terms of AUC$_{(0\text{-infinity})}$, rVIII-SingleChain was not

bioequivalent to Advate. The PK of r-VIII-SingleChain was comparable following single and repeat dosing (3 to 6 months after the initial PK). The PK parameters of rVIII-SingleChain were comparable between non-Japanese and Japanese subjects.

Table 2: Mean (CV%) FVIII PK Parameters for Subjects Dosed at
50 IU/kg ±10% (Part 1) – PK Population

Parameter, unit	ChS		OS	
	Advate 50 IU/kg (n=26)	rVIII-SingleChain 50 IU/kg (n=26)	Advate 50 IU/kg (n=26)	rVIII-SingleChain 50 IU/kg (n=26)
C_{max} (IU/dL)	138 (14.8)	114 (14.7)	105 (20.2)	55.5 (13.9)
IR (IU/dL)/(IU/kg)	2.76 (15.0)	2.27 (14.7)	2.10 (20.3)	1.08 (15.5)
$AUC_{(0-last)}$ (IU*h/dL)	1770 (32.9)	2030 (27.8)	2000 (31.3)	1260 (28.6)
$AUC_{(0-INF)}$ (IU*h/dL)	1850 (34.6)	2130 (29.8)	2100 (34.3)	1340 (33.2)
$T_{1/2}$ (h)	13.4 (33.0)	14.7 (25.4)	14.0 (31.5)	15.5 (31.9)
CL (mL/h/kg)	3.05 (37.1)	2.55 (28.9)	2.69 (37.4)	4.13 (31.6)
Vss (mL/kg)	47.9 (18.9)	49.3 (13.5)	50.4 (20.3)	88.8 (14.0)
MRT (h)	17.3 (32.3)	20.6 (26.5)	20.4 (31.0)	23.3 (31.9)

IR, incremental recovery; C_{max}, maximum observed concentration/activity; AUC_{0-last}, area under the activity-time curve from zero (time of drug administration) to the time of last measurable (positive) activity; $AUC_{0-\infty}$, area under the activity-time curve from zero (time of drug administration) to infinity with extrapolation of the terminal phase; t½, terminal elimination half-life of the compound; CL, total plasma clearance; Vss, volume of distribution at steady state; MRT, mean residence time.

All parameters (except C_{max} and IR) are predose-uncorrected.

Table 3: Mean (CV%) of FVIII Initial and Repeat PK Parameters for All Subjects
with PK Data (Part 3) – PK Population

Parameter, unit	ChS		OS	
	Initial (n=64)	Repeat (n=30)	Initial (n=64)	Repeat (n=30)
C_{max} (IU/dL)	99.9 (19.9)	108 (17.2)	46.3 (19.1)	47.2 (19.3)
IR (IU/dL)/(IU/kg)	1.85 (21.8)	1.99 (17.7)	0.839 (21.9)	0.867 (20.8)
$AUC_{(0-last)}$ (IU*h/dL)	1780 (34.5)	1850 (33.0)	1070 (38.2)	1040 (35.1)
$AUC_{(0-INF)}$ (IU*h/dL)	1830 (34.9)	1880 (34.5)	1090 (39.8)	1050 (36.1)
$T_{1/2}$ (h)	14.1 (27.1)	12.9 (29.4)	14.5 (28.4)	14.1 (26.8)
CL (mL/h/kg)	3.15 (38.2)	3.05 (36.0)	5.56 (46.0)	5.46 (35.5)
Vss (mL/kg)	59.5 (23.9)	53.1 (16.4)	109 (22.8)	105 (17.6)
MRT (h)	20.3 (26.4)	18.9 (28.5)	22.0 (29.1)	20.7 (27.4)

IR, incremental recovery; C_{max}, maximum observed concentration/activity; AUC_{0-last}, area under the activity-time curve from zero (time of drug administration) to the time of last measurable (positive) activity; $AUC_{0-\infty}$, area under the activity-time curve from zero (time of drug administration) to infinity with extrapolation of the terminal phase; t½, terminal elimination half-life of the compound; CL, total plasma clearance; Vss, volume of distribution at steady state; MRT, mean residence time.

All parameters (except C_{max} and IR) are predose-uncorrected.

Table 4: Summary of FVIII Pharmacokinetic Parameters Comparing Subjects by Age Group after Initial and Repeat Injections of rVIII-SingleChain for Subjects Dosed at 50 IU/kg (ChS Assay)

Parameter, unit	Mean (CV%)			
	≥ 12 to < 18 years		≥ 18 to ≤ 65 years	
	Initial PK (n=10)	Repeat PK (n=5)	Initial PK (n=81)	Repeat PK (n=25)
C_{max}, IU/dL	89.7 (24.8)	102 (8.27)	106 (18.1)	109 (18.3)
IR, (IU/dL)/(IU/kg)	1.69 (24.8)	1.86 (13.7)	2.00 (20.8)	2.02 (18.2)
$AUC_{(0-last)}$ IU*h/dL	1480 (36.5)	1980 (18.2)	1890 (32.0)	1800 (36.5)
$AUC_{(0-inf)}$, IU*h/dL	1540 (36.5)	2030 (18.9)	1960 (33.1)	1850 (37.4)
$T_{1/2}$, h	14.3 (33.3)	14.9 (26.4)	14.2 (26.0)	12.5 (29.7)
CL, mL/h/kg	3.80 (46.9)	2.52 (19.5)	2.90 (34.4)	3.15 (36.8)
V_{ss}, mL/kg	68.5 (29.9)	54.6 (16.1)	55.2 (20.8)	52.9 (16.8)
MRT, h	20.0 (32.2)	22.3 (25.2)	20.4 (25.8)	18.2 (28.5)

AUC_{0-last}, area under the activity-time curve from zero (time of drug administration) to the time of last measurable (positive) activity; $AUC_{0-\infty}$ area under the activity-time curve from zero (time of drug administration) to infinity with extrapolation of the terminal phase; CL, clearance; C_{max}, observed maximum plasma concentration; ChS, chromogenic substrate; CV, coefficient of variation; IR, incremental recovery; MRT, mean residence time; n, number of subjects within a specific age group and study part; $t_{1/2}$, half-life; V_{ss}, volume of distribution at steady state.

All parameters (except C_{max} and IR) are predose-uncorrected.

Table 5: Summary of FVIII Pharmacokinetic Parameters Comparing Subjects by Age Group after Initial and Repeat Injections of rVIII-SingleChain for Subjects Dosed at 50 IU/kg (OS Assay)

Parameter, unit	Mean (CV%)			
	≥ 12 to < 18 years		≥ 18 to ≤ 65 years	
	Initial PK (n=10)	Repeat PK (n=5)	Initial PK (n=81)	Repeat PK (n=25)
C_{max}, IU/dL	44.8 (23.8)	50.7 (24.2)	49.3 (18.8)	46.5 (18.2)
IR, (IU/dL)/(IU/kg)	0.806 (25.3)	0.933 (26.0)	0.919 (22.5)	0.853 (19.7)
$AUC_{(0-last)}$, IU*h/dL	911 (35.7)	1200 (8.78)	1140 (35.4)	1000 (38.8)
$AUC_{(0-inf)}$, IU*h/dL	928 (36.7)	1220 (10.0)	1180 (38.3)	1020 (39.9)
$T_{1/2}$, h	13.3 (32.5)	14.9 (31.0)	14.9 (29.1)	13.9 (26.4)
CL, mL/h/kg	6.27 (47.5)	4.11 (10.6)	5.04 (44.6)	5.73 (35.1)
V_{ss}, mL/kg	114 (19.4)	95.5 (19.6)	102 (23.4)	107 (17.0)
MRT, h	20.5 (34.7)	23.6 (25.9)	22.5 (29.5)	20.2 (27.5)

AUC_{0-last}, area under the activity-time curve from zero (time of drug administration) to the time of last measurable (positive) activity; $AUC_{0-\infty}$ area under the activity-time curve from zero (time of drug administration) to infinity with extrapolation of the terminal phase; CL, clearance; C_{max}, observed maximum plasma concentration; CV, coefficient of variation; IR, incremental recovery; MRT, mean residence time; n, number of subjects within a specific age group and study part; OS, one-stage clotting; $t_{1/2}$, half-life; V_{ss}, volume of distribution at steady state.

Table 6: Summary of FVIII Pharmacokinetic Parameters Comparing Non-Japanese and Japanese Subjects After Initial and Repeat Injections of rVIII-SingleChain for all Subjects Dosed at 50 IU/kg (ChS Assay)

Parameter (unit)	Mean (CV%)			
	Initial PK		Repeat PK	
	Non-Japanese (n=54)	Japanese (n=10)	Non-Japanese (n=22)	Japanese (n=8)
Predose-uncorrected				
C_{max} (IU/dL)	98.2 (20.9)	109 (11.9)	109 (17.9)	103 (15.3)
IR (IU/dL)/(IU/kg)	1.81 (23.1)	2.07 (11.2)	2.0 (19.4)	1.97 (13.1)
$AUC_{(0-last)}$ (IU*h/dL)	1740 (37.3)	2000 (15.7)	1840 (36.9)	1900 (22.0)
AUC_{inf} (IU*h/dL)	1790 (37.7)	2060 (15.9)	1860 (38.6)	1940 (22.9)
$T_{1/2}$ (h)	13.7 (25.7)	16.4 (29.0)	12.2 (28.6)	14.8 (27.9)
CL (mL/h/kg)	3.28 (38.6)	2.49 (17.1)	3.16 (37.1)	2.73 (30.9)
V_{ss} (mL/kg)	60.2 (24.4)	55.7 (20.5)	52.6 (17.1)	54.7 (15.4)
MRT (h)	19.9 (26.1)	23.0 (25.4)	18.0 (27.7)	21.3 (28.3)

IR, incremental recovery; C_{max}, maximum observed concentration/activity; AUC_{0-last}, area under the activity-time curve from zero (time of drug administration) to the time of last measurable (positive) activity; $AUC_{0-\infty}$, area under the activity-time curve from zero (time of drug administration) to infinity with extrapolation of the terminal phase; t½, terminal elimination half-life of the compound; CL, total plasma clearance; Vz, volume of distribution; MRT, mean residence time.

All parameters (except C_{max} and IR) are predose-uncorrected.

Table 7: Summary of FVIII Pharmacokinetic Parameters Comparing Non-Japanese and Japanese Subjects After Initial and Repeat Injections of rVIII-SingleChain for all Subjects Dosed at 50 IU/kg (OS Assay)

Parameter (unit)	Mean (CV%)			
	Initial PK		Repeat PK	
	Non-Japanese (n=54)	Japanese (n=10)	Non-Japanese (n=22)	Japanese (n=8)
Predose-uncorrected				
C_{max} (IU/dL)	46.2 (20.5)	46.4 (9.32)	49.0 (20.1)	42.4 (10.0)
IR (IU/dL)/(IU/kg)	0.837 (22.5)	0.853 (19.2)	0.889 (23.0)	0.810 (9.59)
$AUC_{(0-last)}$ (IU*h/dL)	1040 (41.2)	1180 (19.7)	1040 (37.9)	1010 (27.3)
$AUC_{0-\infty}$ (IU*h/dL)	1070 (42.8)	1210 (20.8)	1060 (39.1)	1040 (28.3)
$T_{1/2}$ (h)	14.2 (29.1)	16.0 (23.9)	13.3 (23.0)	16.2 (29.9)
CL (mL/h/kg)	5.79 (46.4)	4.30 (23.5)	5.52 (35.0)	5.29 (39.5)
V_{ss} (mL/kg)	110 (23.8)	103 (14.5)	101 (17.8)	116 (13.7)
MRT (h)	21.4 (29.3)	25.1 (25.9)	19.6 (25.0)	23.9 (28.5)

IR, incremental recovery; C_{max}, maximum observed concentration/activity; AUC_{0-last}, area under the activity-time curve from zero (time of drug administration) to the time of last measurable (positive) activity; $AUC_{0-\infty}$, area under the activity-time curve from zero (time of drug administration) to infinity with extrapolation of the terminal phase; t½, terminal elimination half-life of the compound; CL, total plasma clearance; Vz, volume of distribution; MRT, mean residence time.

All parameters (except C_{max} and IR) are predose-uncorrected.

Table 8: Summary of FVIII Pharmacokinetic Parameters Comparing by Formulation Strength Following Initial Administration of 50 IU/kg rVIII SingleChain

Parameter (unit)	PK Parameter Summary	
	rVIII-SingleChain 3000 IU (n=9)	rVIII-SingleChain 250 IU (n=8)
C_{max} (IU/dL)	104 (23.1)	106 (9.60)
IR (IU/dL)/(IU/kg)	1.95 (24.3)	2.03 (10.2)
$AUC_{(0-last)}$ (IU*h/dL)	1990 (40.1)	1850 (20.8)
$AUC_{(0-INF)}$ (IU*h/dL)	2050 (41.2)	1910 (20.8)
$T_{1/2}$ (h)	14.3 (29.0)	16.0 (31.8)
CL (mL/h/kg)	2.76 (35.9)	2.74 (23.5)
Vss (mL/kg)	53.9 (22.0)	58.5 (19.5)
MRT (h)	21.0 (28.9)	22.2 (27.0)

IR, incremental recovery; C_{max}, maximum observed concentration/activity; AUC_{0-last}, area under the activity-time curve from zero (time of drug administration) to the time of last measurable (positive) activity; $AUC_{0-\infty}$, area under the activity-time curve from zero (time of drug administration) to infinity with extrapolation of the terminal phase; $t_{1/2}$, terminal elimination half-life of the compound; CL, total plasma clearance; Vss, volume of distribution at steady state; MRT, mean residence time.

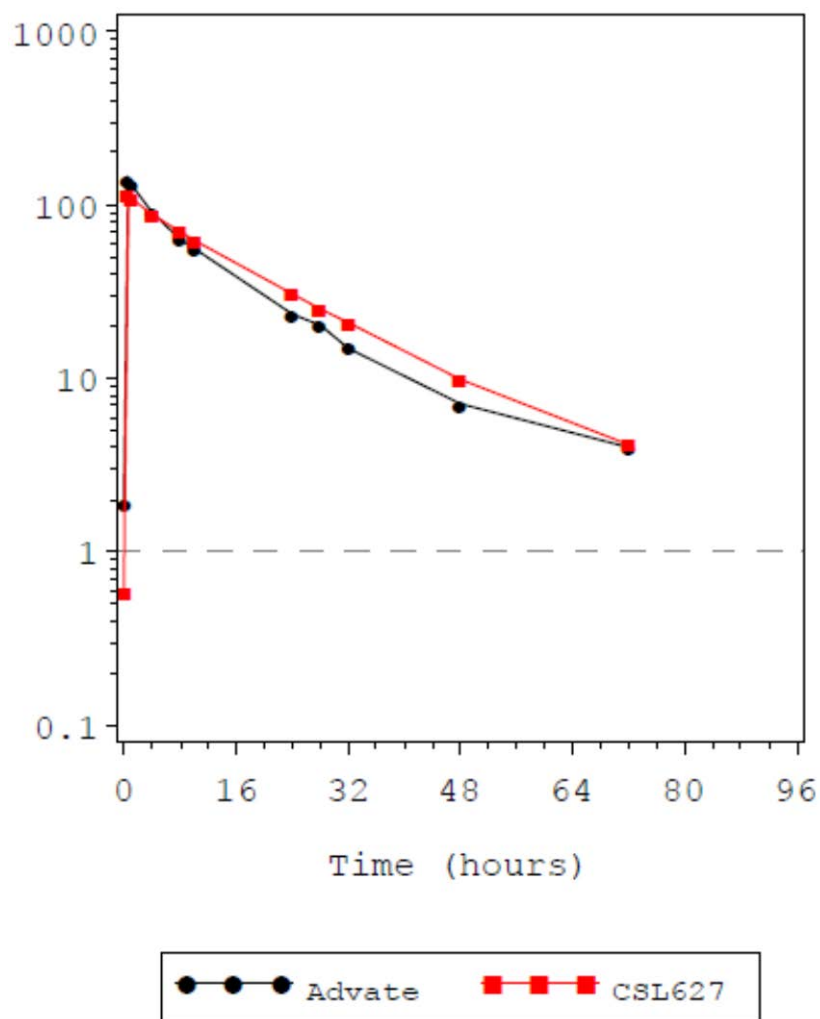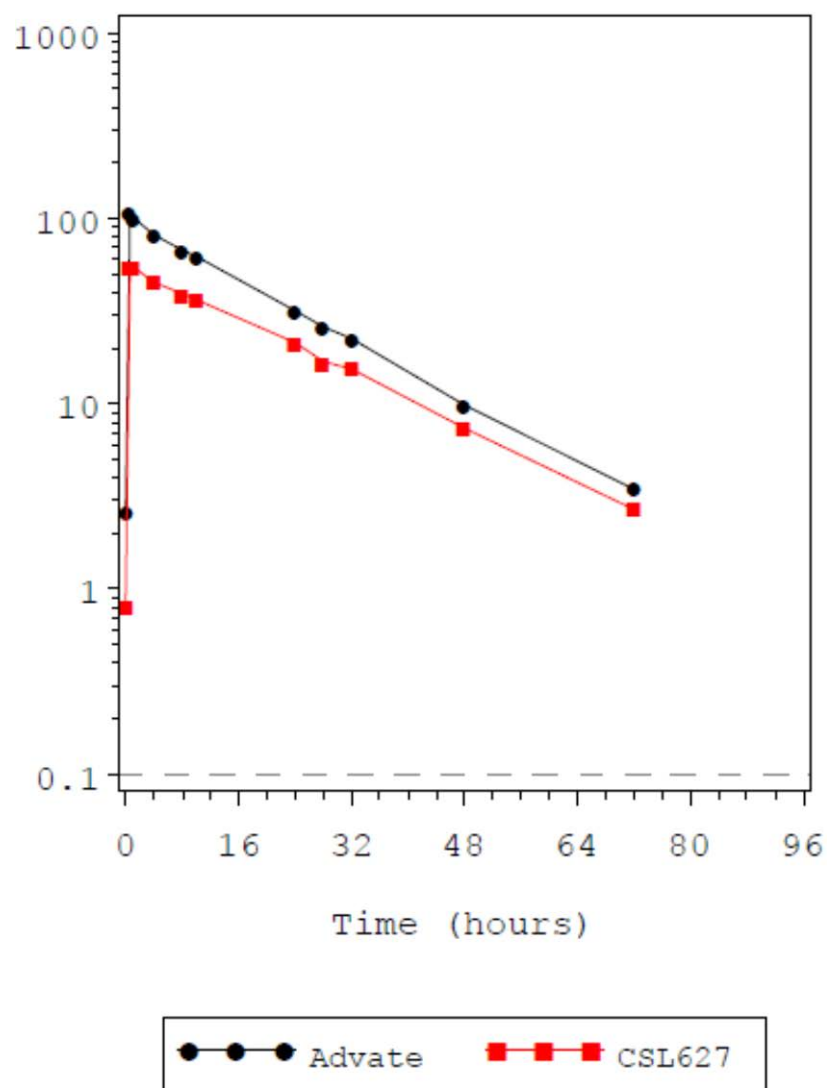All parameters (except C_{max} and IR) are predose-uncorrected.

Figure 1: Mean predose-uncorrected FVIII activity-time profiles after administration of single injection of 50 IU/kg rVIII-SingleChain and 50 IU/kg Advate for subjects dosed at 50 IU/kg ±10% - Part 1 (ChS Assay)

Figure 2: Mean predose-uncorrected FVIII activity-time profiles after administration of single injection of 50 IU/kg rVIII-SingleChain and 50 IU/kg Advate for subjects dosed at 50 IU/kg ±10% - Part 1 (OS Assay)

Study #2

Study Title: A Phase III open-label pharmacokinetic, efficacy and safety study of rVIII-Single Chain in a pediatric population with severe Hemophilia A (CSL627-3002).

This is a multicenter, open-label study to assess the efficacy, safety, and PK of rVIII-SingleChain in pediatric patients with severe hemophilia A. The PK study of rVIII-SingleChain was conducted in children 0-<6 (1 to 5 years; n =20) and >6-<12 (6-<11; n = 19) years of age. The children received 50 IU/kg FVIII dose. Blood samples were taken at time 0, 1, 5, 10, 24, and 48 hours post-dose. The FVIII activity was measured by chromogenic as well as one-stage method. The PK parameters of rVIII-SingleChain for two age groups by two assay methods are summarized in Table 1. FVIII concentration-time profiles are shown in Figures 1-2.

Table 1: PK parameters of rVIII-SingleChain for two age groups (50 IU/kg dose)

| | Mean (CV%) | | | |
| | 0 to < 6 years (N = 19) | | > 6 to < 12 years (N = 19) | |
Parameter, unit	ChS	OS	ChS	OS
C_{max}, IU/dL	80.2 (21.2)	45.0 (81.8)	83.5 (19.5)	42.3 (20.1)
IR, (IU/dL)/(IU/kg)	1.61 (21.4)	0.893 (82.8)	1.66 (19.7)	0.841 (20.3)
AUC_t, IU*h/dL	1020 (28.4)	564 (44.7)	1090 (26.4)	635 (32.3)
AUC_{inf}, IU*h/dL	1090 (31.4)	632 (59.3)	1170 (26.3)	683 (33.2)
$t_{1/2}$, h	10.5 (28.6)	11.1 (48.8)	10.2 (19.4)	10.3 (26.6)
CL, mL/h/kg	5.01 (30.3)	9.73 (45.4)	4.63 (29.5)	8.44 (42.4)
V_{ss}, mL/kg	70.8 (12.1)	134 (27.5)	67.1 (22.3)	121 (21.7)
MRT, h	12.6 (24.1)	12.5 (31.2)	12.3 (16.8)	12.8 (17.5)

Abbreviations: AUC_t, area under the activity / concentration curve from time point zero to the last quantifiable time point; AUC_{inf}, area under the activity / concentration curve extrapolated to infinity; ChS, chromogenic substrate (assay); CL, clearance; CV, coefficient of variation; C_{max}, maximum observed concentration / activity; FVIII, factor VIII; IR, incremental recovery; IU, International Units; MRT, mean residence time; N, total number of subjects; OS, one-stage clotting (assay); PK, pharmacokinetic(s); $t_{1/2}$, half-life; V_{ss}, volume of distribution at steady-state.

Note:
[1] All parameters (except C_{max} and IR) are predose-uncorrected.
Source: Table 14.2.12.4

In children 1-<6 years of age, by chromogenic assay, the in-vivo recovery (IVR), half-life and clearance were 1.61 (IU/dL)/(IU/kg) (%CV = 21.4), 10.5 hours (%CV = 28.6), and 5 mL/hour per kg (%CV = 30.3), respectively. In children 1-<6 years of age, by one-stage assay, the in-vivo recovery, half-life and clearance were 0.9 (IU/dL)/(IU/kg) (%CV = 82.8), 11.1 hours (%CV = 48.8), and 9.73 mL/hour per kg (%CV = 45.4), respectively.

In children >6-<12 years of age, by chromogenic assay, the in-vivo recovery, half-life and clearance were 1.66 (IU/dL)/(IU/kg) (%CV = 19.7), 10.2 hours (%CV = 19.4), and 4.63 mL/hour

per kg (%CV = 29.5), respectively. In children >6 <12 years of age, by one-stage assay, the in-vivo recovery, half-life and clearance were 0.84 (IU/dL)/(IU/kg) (%CV = 20.3), 10.3 hours (%CV = 26.6), and 8.44 mL/hour per kg (%CV = 42.4), respectively.

The results of the study indicated that the aforementioned PK parameters were comparable between children 1-<6 and >6 <12 years of age by both assay methods (comparison within the assay method). The chromogenic assay provided lower clearance and higher IVR than one-stage assay in both age groups. On the other hand, the half-life of FVIII was comparable by both assay methods between the two age groups. The one-stage assay method produced higher %CV than chromogenic assay in children 1-<6 years of age.

Comparison of clearance and half-life across age groups:

Chromogenic Assay:
The clearance and half-life of FVIII in adults (>18 years of age) were 2.55 mL/hr per kg and 14.7 hours, respectively. In children 1-<6 years of age, clearance and half-life were 5 mL/hour per kg and 10.5 hours, respectively. The clearance of FVIII in this age group was approximately 2-fold higher and half-life was approximately 4 hours shorter than adults.

In children >6-<12 years of age, clearance and half-life were 4.6 mL/hour per kg and 10.2 hours, respectively. The clearance of FVIII in this age group was approximately 1.8-fold higher and half-life was approximately 4 hours shorter than adults.

One-stage Assay:
The clearance and half-life of FVIII in adults (>18 years of age) were 4.1 mL/hr per kg and 15.5 hours, respectively. In children 1-<6 years of age, clearance and half-life were 9.7 mL/hour per kg and 11.1 hours, respectively. The clearance of FVIII in this age group was approximately 2.4-fold higher and half-life was approximately 4 hours shorter than adults.

In children >6-<12 years of age, clearance and half-life were 8.4 mL/hour per kg and 10.3 hours, respectively. The clearance of FVIII in this age group was approximately 2-fold higher and half-life was approximately 5 hours shorter than adults.

Conclusions: The body weight adjusted clearance of rVIII-SingleChain in children <12 years is higher than adults by 1.7 to 2-fold. The half-life of rVIII-SingleChain is shorter than adults in this age group by 4 to 5 hours. Based on these observations, higher doses will be needed in children.

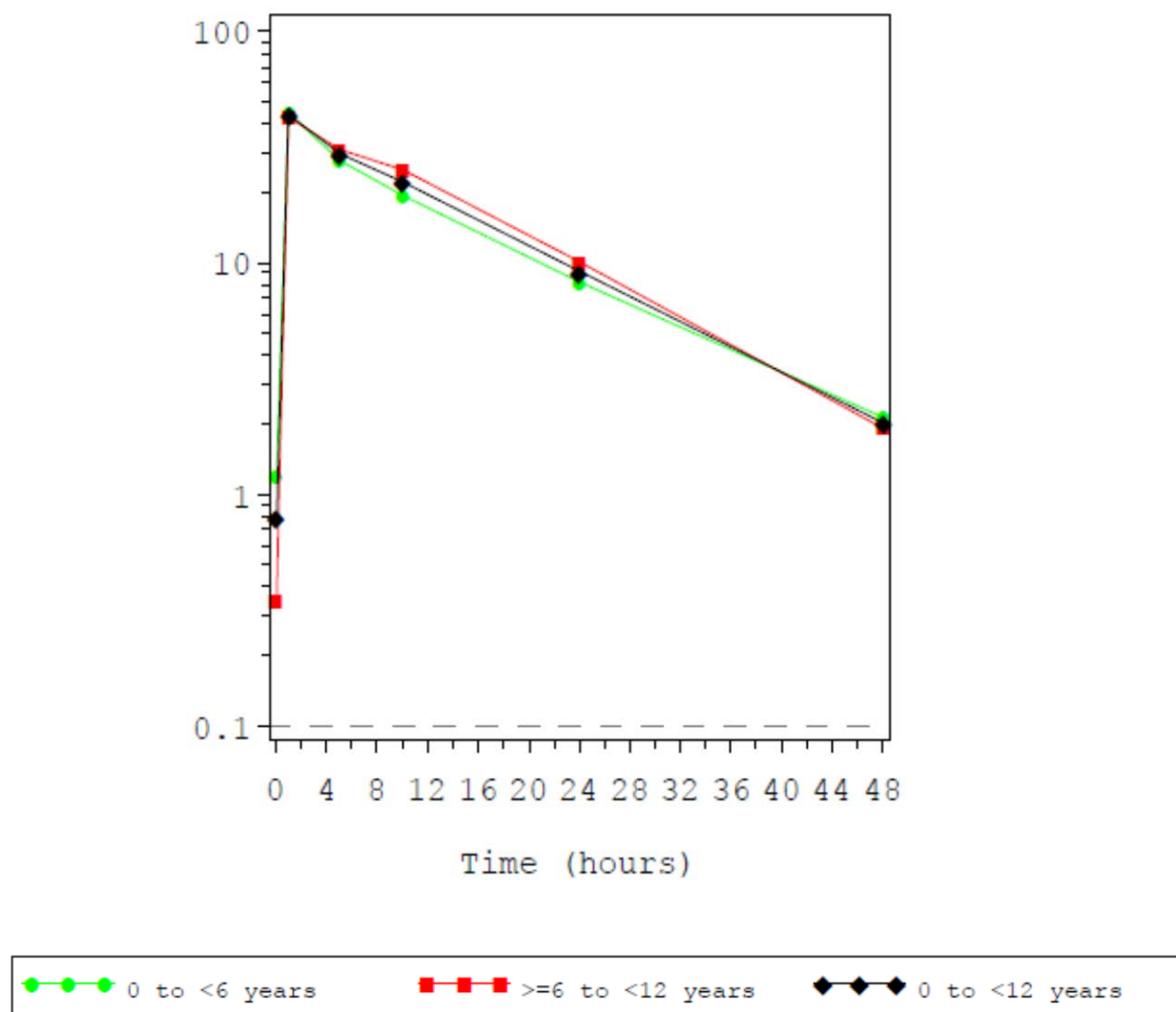**Figure 1: Mean predose-uncorrected FVIII activities by age (OS assay)**

Figure 2: Mean predose-uncorrected FVIII activities by age (Chs assay)