CENTER FOR DRUG EVALUATION AND RESEARCH

APPLICATION NUMBER:

761136Orig2s000

CLINICAL PHARMACOLOGY AND BIOPHARMACEUTICS REVIEW(S)

Office of Clinical Pharmacology Review

NDA or BLA Number	BLA 761136
Link to EDR	\\CDSESUB1\evsprod\BLA761136\0001
Submission Date	04/04/2019
Submission Type	*Standard Review*
Brand Name	REBLOZYL®
Generic Name	Luspatercept-aamt
Dosage Form and Strength	25 mg or 75 mg lyophilized powder in single-dose vials
Route of Administration	Subcutaneous injection
Proposed Indication	Adult patients with very low- to intermediate-risk myelodysplastic syndromes (MDS)-associated anemia who have ring sideroblasts and require red blood cell (RBC) transfusions
Proposed Dosing Regimen	• Starting dose of 1.0 mg/kg once every 3 weeks as a subcutaneous injection. • If a patient is not RBC transfusion-free after at least 2 consecutive doses (6 weeks) at the 1.0 mg/kg starting dose, increase the REBLOZYL dose to 1.33 mg/kg. • If a patient is not RBC transfusion-free after at least 2 consecutive doses (6 weeks) at the 1.33 mg/kg dose, increase the REBLOZYL dose to 1.75 mg/kg. • Do not increase the dose beyond the maximum dose of 1.75 mg/kg.
Applicant	Celgene Corporation
Associated IND	IND 112562
OCP Review Team	*Lili Pan, Ph.D.* *Liang Li, Ph.D.* *Lian Ma, Ph.D.* *Guoxiang (George) Shen, Ph.D.*
OCP Final Signatory	*Brian Booth, Ph.D.*

Table of Contents

LIST OF TABLES

LIST OF FIGURES

1. EXECUTIVE SUMMARY

Luspatercept is a recombinant fusion protein that binds select TGF-β superfamily ligands and inhibits Smad2/3 signaling, resulting in erythroid maturation. The proposed indication for luspatecept in this submission, is the treatment of adult patients with very low- to intermediate-risk myelodysplastic syndromes (MDS)-associated anemia who have ring sideroblasts and require red blood cell (RBC) transfusions.

In a randomized, open-label, multicenter study (ACE-536-MDS-001) that included 229 patients with lower-risk MDS, patients were randomized 2:1 to receive either luspatercept or placebo in 21-day cycles. The response rate in the luspatercept arm was 37.9% (95% CI: 30.2-46.1%) compared to a response rate of 13.2% (95% CI: 6.5-22.9%) in the placebo arm, with regard to the primary endpoint as RBC transfusion free ≥ 8 weeks during Weeks 1-24.

The proposed titration-to-response dosing regimen (1.0 to 1.33 to 1.75 mg/kg), was found acceptable with both primary and secondary efficacy endpoints achieved. Exposure-safety analyses identified a generally flat relationship between luspatercept exposure and the probability of Grade ≥3 treatment-emergent adverse events (TEAEs). Dose-neoplasm analysis indicated that there was no clear relationship between incidence rate of disease progression/second primary malignancy and luspatercept dose/drug exposure.

Population PK analysis suggested that no dose modification is needed for specific populations of age, sex, race, mild or moderate renal impairment, mild to severe hepatic impairment, and baseline disease characteristics. These factors were not found to be clinically significant covariates on luspatercept PK.

A total of 8.9% (23/260) patients tested positive for treatment-emergent anti-luspatercept antibodies, including 3.8% (10/260) patients who had neutralizing antibodies. Luspatercept serum concentration tended to decrease in the presence of neutralizing antibodies. There was no discernible effect of ADAs on efficacy or safety.

1.1 Recommendations

The Office of Clinical Pharmacology has reviewed the information contained in BLA 761136. There are no Clinical Pharmacology related issues that would preclude the approval of this current BLA. The key review issues with specific recommendations/comments are summarized below:

Review Issues	Recommendations and Comments
Evidence of effectiveness	A randomized, double-blind, placebo-controlled Phase 3 Study ACE-536-MDS-001 provides primary evidence. Refer to Statistical Review for details.
General Dosing instructions	• The recommended starting dose of REBLOYZL is 1.0 mg/kg once every 3 weeks by subcutaneous (SC) injection. • If a patient is not RBC transfusion-free after at least 2 consecutive doses (6 weeks) at the 1.0 mg/kg starting dose, increase the REBLOZYL dose to 1.33 mg/kg. • If a patient is not RBC transfusion-free after at least 2 consecutive doses (6 weeks) at the 1.33 mg/kg dose, increase the REBLOZYL dose to 1.75 mg/kg. • Do not increase the dose beyond the maximum dose of 1.75 mg/kg. • Patients must have their Hgb assessed and have results available prior to each administration. If an RBC transfusion occurred prior to dosing, the pre-transfusion Hgb must be considered for dosing purposes. • If the pre-dose Hgb is greater than or equal to 11.5 g/dL and the Hgb level is not influenced by recent transfusion, delay dosing until the Hgb is less than or equal to 11.0 g/dL.
Dosing in patient subgroups (intrinsic and extrinsic factors)	No dose modification is needed for specific populations of age, sex, race, mild or moderate renal impairment, mild to severe hepatic impairment, and baseline disease characteristics. These factors were not found to be clinically significant covariates on luspatercept PK. (Section 2.2)

1.2 Post-Marketing Requirements and Commitments

None.

2. SUMMARY OF CLINICAL PHARMACOLOGY ASSESSMENT

2.1 Pharmacology and Clinical Pharmacokinetics

Luspatercept is a recombinant fusion protein that binds select TGF-β superfamily ligands. By binding to specific endogenous ligands (e.g., GDF-11, activin B), luspatercept inhibits Smad2/3 signaling, resulting in erythroid maturation through differentiation of late-stage erythroid precursors (normoblasts) in the bone marrow. Smad2/3 signaling is abnormally high in disease models characterized by ineffective erythropoiesis, e.g., MDS.

Following SC administration of multiple doses of luspatercept every 3 weeks (Q3W) in patients with MDS, luspatercept drug exposures (i.e., C_{max} & AUC) in serum increased proportionally to dose from 0.125 to 1.75 mg/kg. Following repeat dosing of luspatercept with the recommended Q3W dosing schedule, steady-state exposure was reached after 3 doses with an accumulation ratio of 1.5 for the trough concentration. The geometric mean (%CV) was 9.7 L (26.5%) for apparent volume of distribution (Vd/F), 13 days (31.6%) for terminal half-life ($t_{1/2}$), and 0.52 L/day (41.2%) for apparent clearance (CL/F).

In the Phase 3 trial ACE-536-MDS-001, among 153 patients who were treated with REBLOYZL at the recommended dosing regimen, 11 patients (7.2%) tested positive for treatment-emergent anti-drug antibodies (ADAs), including 5 patients (3.3%) who developed neutralizing antibodies; among 76 patients who were treated with placebo, 3 patients (4.0%) tested positive for treatment-emergent ADAs, including 2 patients (2.6%) developed neutralizing antibodies. Luspatercept serum concentration tended to decrease in the presence of neutralizing antibodies. No discernible efficacy or safety difference were notified.

2.2 Dosing and Therapeutic Individualization

2.2.1 General dosing

The Applicant's proposed starting dose of luspatercept is 1.0 mg/kg once every 3 weeks administered via SC injection. The dose may be increased to 1.33 mg/kg and then to 1.75 mg/kg during treatment if the patient is not RBC transfusion-free at the prior dose level for at least two consecutive treatment cycles (6 weeks). If the patient has pre-dose Hgb ≥ 11.5 g/dL and the Hgb level is not influenced by recent transfusion, delay dosing until Hgb ≤ 11.0 g/dL. This proposed dosing regimen appears to be acceptable from a Clinical Pharmacology perspective.

2.2.2 Therapeutic individualization

No intrinsic or extrinsic factors that would require adjustment of the proposed dosing regimen have been identified.

2.3 Outstanding Issues

There are no outstanding Clinical Pharmacology related issues for this cycle.

2.4 Summary of Labeling Recommendations

Labeling recommendations are generally adequate from a clinical pharmacology perspective.

3. COMPREHENSIVE CLINICAL PHARMACOLOGY REVIEW

3.1 Overview of the Product and Regulatory Background

Luspatercept was previously investigated under IND 112562. Luspatercept received Orphan Drug Designation in December 2012 and Fast Track Designation in October 2015 for treatment of anemia in lower-risk MDS. In July 2015, the proposed study design for registrational trial ACE-536-MDS-001 was discussed between Celgene and FDA.

In November 2019, luspatercept received approval for treatment of anemia in adult patients with beta thalassemia who require regular red blood cell (RBC) transfusion.

3.2 General Pharmacology and Pharmacokinetic Characteristics

Pharmacology	
Mechanism of Action	Luspatercept is a recombinant fusion protein that binds select TGF-β superfamily ligands and inhibits Smad2/3 signaling, resulting in erythroid maturation.
Active Moieties	Luspatercept
QT Prolongation	At a dose 0.125 to 1.75 times the approved recommended dosage, the safety QTc data do not suggest any off-target effects for QTc prolongation; the incidence of patients with QTc categorical outliers (e.g., QTc > 500 ms or increase in QTc > 60 ms) is similar between placebo and luspatercept arms. See BLA 761136 DARRTS CONSULT REV-QTIRT-01 dated 06/20/2019 by ZHENG for details.
General Information	
Bioanalysis	Luspatercept was measured using validated Enzyme Linked Immunosorbent Assay (ELISA) method.
Drug exposure after first dose	Following SC dose of 1.0 mg/kg, the geometric mean C_{max} of 6.44 [CV%: 16.6] µg/mL was reached at day 5.55.
Drug total exposure at steady state following the therapeutic dosing regimen	Following multiple SC doses of 1.0 mg/kg Q3W, the steady state geometric mean $C_{max,ss}$ and AUC_{ss} were 9.29 [CV%: 30.0] µg/mL and 148 [CV%: 37.5] day·µg/mL; following multiple SC doses of 1.33 mg/kg Q3W, the steady state geometric mean $C_{max,ss}$ and AUC_{ss} were 12.4 [CV%: 30.0] µg/mL and 196 [CV%: 37.5] day·µg/mL; following multiple SC doses of 1.75 mg/kg Q3W, the steady state geometric mean $C_{max,ss}$ and AUC_{ss} were 16.3 [CV%: 30.0] µg/mL and 258 [CV%: 37.5] day·µg/mL.
Minimal effective dose or exposure	1.0 mg/kg administered via SC injection once every 3 weeks.
Dose Proportionality	Luspatercept serum exposure (AUC_{ss} and C_{max}) increased approximately dose-proportionally with SC doses from 0.125 to 1.75 mg/kg.
Accumulation	The accumulation ratio was approximately 1.5-fold. Steady state of exposure was reached after 3 doses Q3W.
Variability	The %CV for C_{max} was 20.5% after the first dose and 29.9% at steady state. The %CV for AUC_{ss} was 38.3%.
Immunogenicity	A total of 8.9% (23/260) patients who were treated with luspatercept tested positive for treatment-emergent anti-luspatercept antibodies,

	including 3.8% (10/260) who had neutralizing antibodies. A total of 4.0% (3/76) patients who were treated with placebo tested positive for treatment-emergent ADAs, including 2.6% (2/76) who developed neutralizing antibodies. Luspatercept serum concentration tended to decrease in the presence of neutralizing antibodies. There was no discernible effect of ADAs on efficacy or safety.
Distribution	
Volume of Distribution	The apparent volume of distribution (%CV) was 9.8 L (26.5%).
Plasma Protein Binding	Not evaluated. As a fusion protein with a molecular weight of 76 kDa, luspatercept is not expected to bind to plasma proteins.
Blood to Plasma Ratio	Not evaluated.
Substrate transporter	Not evaluated. As a fusion protein, luspatercept is not expected to be a substrate of metabolic transporters.
Elimination	
Clearance	The apparent clearance (%CV) was 0.52 L/day (41.2%).
Mean terminal elimination half-life	The terminal phase half-life (CV%) was 13 days (31.6%).
Metabolism	
Primary metabolic pathway(s)	No evaluated. Luspatercept is expected to be catabolized into amino acids by general protein degradation processes in multiple tissues, and thus its elimination is not dependent on a single organ.
Inhibitor/Inducer	Not evaluated.
Excretion	
Primary excretion pathways (% dose) ± SD	Not evaluated. Luspatercept is not expected to be excreted into urine due to its large molecular mass (76 kDa) that is above the glomerular filtration cut-off threshold (~65 kDa).

3.3 Clinical Pharmacology Review Questions

3.3.1 Does the clinical pharmacology program provide supportive evidence of effectiveness?

Yes, the clinical pharmacology program provides supportive evidence of effectiveness. In the PK/PD analysis from the Phase 2 study A536-03, a dose-dependent increase from baseline in Hgb was observed in patients with baseline RBC-T < 4 units/8 weeks, while dose-dependent increase from RBC-T reduction was observed in patients with baseline RBC-T ≥ 4 units/8 weeks. See Section 4.2 Clinical PK/PD and Immunogenicity Assessments for details.

The applicant submitted safety and efficacy results from Study ACE-536-MDS-001 to support approval for the proposed indication in patients with lower-risk MDS associated anemia. Additionally, the applicant submitted data from a Phase 2 study A536-03 to support dose selection and data from Phase 2 study A536-05 as supportive evidence of safety and efficacy of luspatercept (Table 1).

Table 1: Clinical Studies Supporting Luspatercept Treatment in Patients with Lower-Risk MDS associated Anemia

Trial	Design	Luspatercept Regimen	Analysis Population
Registrational Trial			
ACE-536-MDS-001	A Phase 3, double-blind, randomized study: • Treatment Arm: Luspatercept + BSC • Placebo Arm: Placebo + BSC	• Subcutaneous (SC) administration once every 3 weeks (Q3W) • Starting dose: 1.0 mg/kg • Dose escalation to 1.33 mg/kg then 1.75 mg/kg if no RBC transfusion free after 2 consecutive doses	Patients: IPSS-R Very Low, Low, or Intermediate Risk MDS with Ring Sideroblasts Who Require Red Blood Cell Transfusions Treatment Arm: N=153 Placebo Arm: N=76
Supportive Studies			
A536-03	A Phase 2, open-label, ascending dose study of luspatercept	• SC administration Q3W • Dose escalation cohort: 0.125, 0.25, 0.5, 0.75, 1, 1.33, 1.75 mg/kg • Dose expansion cohort: Starting dose 1 mg/kg, may escalate to 13, 1.75 mg/kg	Patients: IPSS-R Very Low, Low, or Intermediate Risk MDS N = 107
A536-05	A Phase 2, open-label, extension study for long-term effects	• SC administration Q3W • Starting dose 1 mg/kg, may escalate to 13, 1.75 mg/kg	Patients: IPSS-R Very Low, Low, or Intermediate Risk MDS N = 70

3.3.2 Is the proposed dosing regimen appropriate for the general patient population for which the indication is being sought?

Yes, the proposed luspatercept starting dose and dose titration scheme is overall supported by PK, efficacy and safety findings in the indicated population.

Dose Selection Rationale for Phase 3

Body weight-based dosing: Body weight was a statistically significant covariate of luspatercept apparent CL/F and Vd/F in the population PK analysis. A total of 100 trials were simulated based on the PK model to compare 3 dosing regimens in 336 patients: a weight-based dosing regimen (1.75 mg/kg), a modified weight-based dose regimen (1.75 mg/kg up to 168 mg), and fixed dose (133 mg). Results predicted that the weight-based dose regimen would perform better than the fixed dosing by decreasing the exposure difference between lighter and heavier patients and the typical patients to within 10%, instead of the 25-30% predicted for the fixed dose.

Q3W dosing schedule: A Q3W dosing schedule is expected to maintain approximately 50% of the peak concentration at the end of a dosing interval as luspatercept has T_{max} ~5.4 days and $t_{1/2}$ ~13 days in patients with lower-risk MDS. Following the Q3W dosing schedule, the mean C_{trough} at steady state (≥ 3.5 µg/mL or ≥ 46 nM) was far above the K_d of luspatercept binding to GDF11 (0.71 nM), or the IC_{50} of luspatercept to inhibit signaling through GDF11 (7.1 ng/mL) in *in-vitro* assays.

Starting dose 1.0 mg/kg and dose titration to 1.33 mg/kg then 1.75 mg/kg:

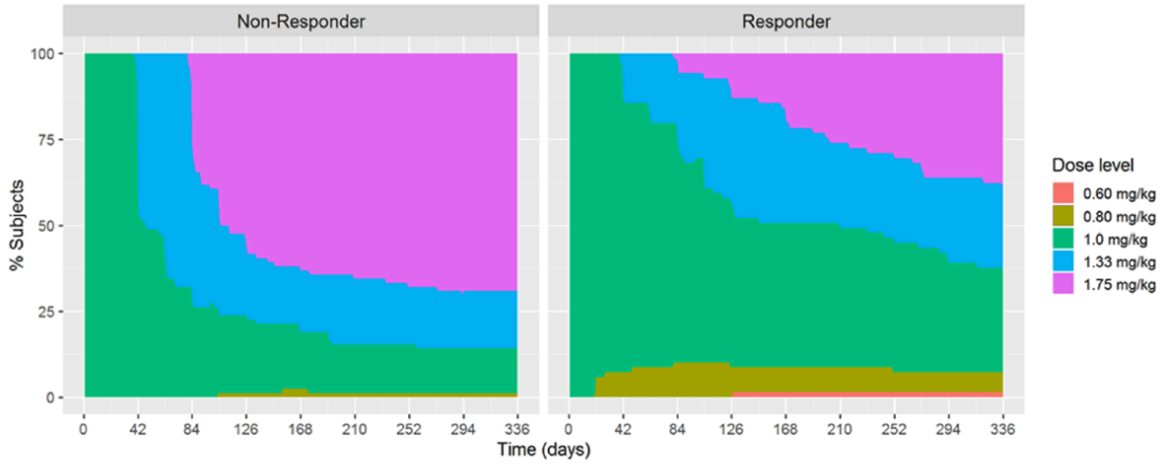
- In the supportive phase 2 study A536-03, higher response rates in Hgb increase ≥ 1.5 g/dL were sustained for ≥ 14 days, and RBC-transfusion freedom was observed at dose ≥ 0.8 mg/kg within the studied dose range of 0.125 - 1.75 mg/kg.
- Luspatercept dose levels up to 1.75 mg/kg were tolerated in patients with lower-risk MDS. In the phase 2 studies, MTD was not reached at 1.75 mg/kg for up to 5 treatment cycles in Study A536-03.

Supportive Evidence from Phase 3

The proposed titration-to-response dosing regimen (1-1.75 mg/kg) was confirmed to be effective in the Phase 3 study ACE-536-MDS-001 with both primary and secondary efficacy endpoints achieved. Refer to Clinical and Stats reviews.

The overall percentages of patients who received 1.0 mg/kg, 1.33 mg/kg, or 1.75 mg/kg as the maximum dose in the Phase 3 study ACE-536-MDS-001 were 23%, 18%, and 59%, respectively. The percentage of dose escalation was greater for non-responders than responders (*Figure 1*).

Figure 1. Dose Distribution over Time Between Responders and Non-Responders in Study ACE-536-MDS-001



Responders are defined as subjects who achieved red blood cell transfusion independence ≥ 8 consecutive weeks during the first 48-week treatment period.
Source: ACE-536-MPK-002, Figure 37.

Dose escalations were more frequently seen in patients who had no splenectomy and had higher baseline EPO (≥ 200 U/L), conditions known to be associated with more resistant anemia or more advanced disease (*Table 2*).

Exposure-safety analyses suggested a generally flat relationship between luspatercept exposure and the probability of Grade ⩾3 TEAEs. Refer to Section 4.4 Exposure-Response Analysis for details. In addition, a dose-neoplasm analysis indicated that there was no clear relationship between incidence rate of disease progression/SPM and luspatercept dose/drug exposure. Refer to Section 4.5 Dose-Neoplasm Analysis for details.

Table 2. Summary of Luspatercept Exposure and Baseline Factors by Maximum Dose Level in Study ACE-536-MDS-001.

Parameter	Statistics	Maximum Dose Level in Week 1-24		
		1 mg/kg (N = 51)	1.33 mg/kg (N = 40)	1.75 mg/kg (N = 62)
Baseline RBC-T burden (units/24 week)	Median (90% PI)	15 (9, 36)	17 (8, 32)	18 (12, 34)
Baseline EPO (U/L)	Median (90% PI)	99 (25, 800)	131 (29, 487)	151 (26, 717)

Source: Report ACE-536-MPK-002, Table 19.

3.3.3 Is an alternative dosing regimen and/or management strategy required for subpopulations based on intrinsic factors?

No. The proposed body weight-based dosing regimen is supported by the Population PK analysis (n = 260), in which both CL/F and Vd/F of luspatercept increased with body weight in patients with lower risk MDS. No statistically meaningful influence on PK of luspatercept was identified for other intrinsic factors such as age (27 - 95 years), sex (38.8% female/61.2% male), race (0.4% Black/82.3% White/), hepatic impairment (31.5% mild, 8.8% moderate and 0.4% severe based on NCI-ODWG criteria), and renal impairment (51.5% mild, and 21.5% moderate based on eGFR), baseline serum erythropoietin (9.8 to 2450 U/L), baseline albumin (31 - 53 g/L), baseline RBC-T burden (0 to 43 units/24 weeks), ring sideroblasts, splenectomy, location of SC injection (i.e., upper arm, thigh, or abdomen), and concurrent iron chelation therapy after the dose was adjusted by body weight. The effect of severe renal impairment (eGFR <30 mL/min/1.73 m^2) is unknown.

Although the effect of age and baseline albumin on CL/F and baseline albumin on Vd/F were statistically significant in the population PK analysis, dose adjustment was not required as their impact on luspatercept exposure was not considered clinically significant, given that dose was proposed to be titrated based on response and the E-R for safety was relatively flat at the dose range of 1.0 m/kg to 1.75 mg/kg. Refer to Section 4.3 Population PK Analysis for further detailed information.

3.3.4 Are there clinically relevant drug-drug interactions and what is the appropriate management strategy?

Since luspatercept is administered via SC injection, food-drug interactions are not anticipated. Drug-drug interactions are not expected with Cytochrome P450 enzymes (CYPs), other metabolizing enzymes, or transporters, as luspatercept is a fusion protein with molecular weight of 76 kDa. Therefore, no drug-drug interaction studies were conducted in vitro or in vivo.

4. APPENDICES

4.1 Summary of Bioanalytical Method Validation and Performance

Bioanalytical Method for Detection of Luspatercept

Bioanalytical methods for the quantitative determination of luspatercept in human serum were developed and validated using an enzyme-linked immunosorbent assay (ELISA). The assay is designed to measure the total luspatercept in serum, with goat polyclonal anti-luspatercept antibody (against the modified extracellular domain [ECD] of human activin receptor type II B [ActRIIB]) as the capture reagent. *Table 3* summarizes the method performance parameters during method validation. The same method has been used to determine luspatercept serum concentrations in all clinical studies. The validation of the method and sample analysis were conducted in compliance with the appropriate regulations in place at the time of execution.

Table 3. Performance parameters for luspatercept during method validation.

Type of assay	**Enzyme linked immunosorbent assay**	
Analytical facility	(b) (4)	
Capture reagent	Goat polyclonal anti-luspatercept antibody	
Detection reagent	Sheep polyclonal anti-human IgG1 HRP and TMB peroxidase substrate	
QC sample concentration	50, 150, 250, 450, and 600 ng/mL, in 100% serum	
Calibration range	50 to 600 ng/mL, in 100% serum	
Minimum dilution ratio	1:20	
Dilution integrity	16000-fold dilution from 1 mg/mL	
Matrix effect	9/10 normal lots met criteria; 9/10 disease (MDS) lots met criteria	
Precision	Inter-assay ≤ 12.0 % CV. Intra-assay ≤ 8.08 % CV.	
Accuracy	Inter-assay -2.33 to 7.60 % RE. Intra-assay -2.35 to 7.62 % RE.	
Stability (in serum)	Ambient temperature	Stable for at least 20.25 hours
	Freeze/thaw at -70°C	Stable for at least 4 freeze/thaw cycles
	Long-term at -20°C	Stable for at least 386 days
	Long-term at -70°C	Stable for at least 1492 days

%CV = % coefficient of variation; % RE = % relative error; HRP = horseradish peroxidase; MDS = myelodysplastic syndromes; TMB = tetramethylbenzidine; QC = quality control.
Source: Report 176680 and addendum 1 (Report 176681).

Source: EDR 2.7.1 Table 3.

Bioanalytical Method for Detection of Anti-Drug Binding and Neutralizing Antibodies to Luspatercept

Detection of ADAs

Bioanalytical methods for the quantitative determination of luspatercept binding ADA in human serum were developed and validated using an electrochemiluminescence (ECL) immunoassay. The method performance parameters during method validation are summarized in *Table 4*. Detection of ADA was

based on the bivalent characteristics of the antibody. Anti-drug antibodies, if present, would form a "bridge" between the luspatercept coating on the plate and the biotinylated luspatercept added, and subsequently detected through addition of streptavidin-sulfotag binding to the biotin domain. Goat polyclonal anti-human ActRIIB antibody (directed against the ECD of human ActRIIB) was used as a positive control. The final confirmation of ADA positive was evaluated via immune-competition with luspatercept, ACE-536-his, and natural ECD of human ActRIIB.

Table 4. Performance parameters for binding ADA assay during method validation.

Type of Assay	**Electrochemiluminescent Immunoassay**	
Analytical facility	(b) (4)	
Positive control (PC)	Goat polyclonal anti-human ActRIIB antibody	
Specificity test	Luspatercept	
	Modified ECD of human ActRIIB (receptor portion of luspatercept)	
	Natural ECD of human ActRIIB (cross reactivity)	
Minimum dilution ratio	1:11.5 (including acidification step)	
QC sample concentration	0.5 and 16 µg/mL, in 100% serum	
Cut point	Screening	1.40 (multiplicative cut point factor)
	Confirmation with luspatercept [a]	36.9% inhibition
	Specificity with ACE-536-His [a]	32.3% inhibition
	Specificity with ActRIIB-ECD [b]	26.8% inhibition
Sensitivity	26.1 to 167 ng/mL PC, in 100% serum [c]	
Precision	Inter-assay ≤ 26.9 % CV. Intra-assay ≤ 4.10 % CV.	
Free drug interference	At 0.5 µg/mL PC	> 0.1 to > 1 µg/mL [d]
	At 16 µg/mL PC	> 10 to > 50 µg/mL [e]

ACE-536-His = histidine-tagged modified ECD of human ActRIIB; ActRIIB = activin receptor type IIB; ActRIIB-ECD = natural ECD of human ActRIIB; CV% = % coefficient of variation; ECD = extracellular domain; PC = positive control; QC = quality control.

[a] Value derived from original report. Subsequent values from addendums differ but all values meet acceptance criteria.

[b] Value derived from Report 177374.

[c] 26.1 to 41.7 ng/mL in addendums 2 and 3 with multiple reagent lot changes, which have been used for all Phase 2 and Phase 3 studies.

[d] > 1 µg/mL in addendums 2 and 3 with multiple reagent lot changes, which have been used for all phase 2 and 3 studies.

[e] > 25 to 50 µg/mL in addendums 2 and 3 with multiple reagent lot changes, which have been used for all phase 2 and 3 studies.

Source: Report 174275 and addendums 1 to 3 (Report 177374, Report 180010, Report 183554).

Source: EDR 2.7.1 Table 4.

Detection of NAbs

Confirmed ADA-positive serum samples were further evaluated in a neutralization assay to assess the ability to interfere with the luspatercept-ligand interaction by a validated ELISA method. This assay was designed to detect neutralizing antibodies (NAb) which bind to immobilized luspatercept and thereby block binding of biotinylated GDF11 to the drug. This method used chicken polyclonal anti-luspatercept antibody (directed against the modified ECD of human ActRIIB) as the positive control (Report #176678).

Serum samples identified as positive for cross-reactivity to the natural ECD of human ActRIIB in the specificity test were also examined in a second validated ELISA-based NAb assay. The second NAb assay was designed to detect NAb which binds to immobilized human ActRIIB-ECD-Fc fusion proteins (ACE-031) and thereby blocks binding of biotinylated activin A to the natural ECD of the ActRIIB-ECD-Fc protein, with mouse anti-ActRIIB monoclonal antibody (directed against ECD of human ActRIIB) as the positive control (Report #207-1001).

Table 5. Performance parameters for neutralizing ADA assay during method validation.

Validation report	176678	207-1001
Type of assay	ELISA	ELISA
Analytical facility		(b) (4)
Analyte	Neutralizing luspatercept antibodies	Neutralizing AcRIIB-ECD-Fc antibodies (cross reactivity)
Positive control (PC)	Chicken polyclonal anti-luspatercept antibody	Mouse monoclonal anti-human ActRIIB antibody
Coated reagent	Luspatercept	ActRIIB-ECD-Fc fusion protein
Binding ligand	Biotinylated GDF11	Biotinylated activin A
Minimum dilution ratio	1:10	1:10
QC sample concentration	15 and 100 µg/mL, in 100% serum	2.5, 6.25, 10, and 20 µg/mL in 100% serum
Cut point	0.952 (multiplicative cut point factor)	14.77 % inhibition
Sensitivity	912 ng/mL PC, in 100% serum	3681 ng/mL PC, in 100% serum
Precision	Inter-assay: ≤ 27.1 % CV	Inter-assay: ≤ 13.8 % CV [a]
	Intra-assay: ≤ 9.29 % CV	Intra-assay: ≤ 16.3 % CV [a]
Free drug interference	At 15 µg/mL PC: ≥ 0.01 µg/mL [b]	At 6.25 µg/mL PC: ≥ 50 µg/mL [c]
	At 100 µg/mL PC: ≥ 0.01 µg/mL [b]	

ActRIIB = activin receptor type IIB; ActRIIB-ECD-Fc = a fusion protein joining the ECD of human ActRIIB to Fc portion of human Ig G; CV% = % coefficient of variation; ECD = extracellular domain; ELISA = enzyme linked immunosorbent assay; NAb = neutralizing antibodies; PC= positive control; QC = quality control.

[a] Results from positive control 2.5 µg/mg are excluded because it fails to meet the inter-assay precision %CV acceptance criteria of ≤ 30%.

[b] Presence of luspatercept as low as 0.01 µg/mL were found to interfere with the assay (reducing positivity signal). However, the overall response remains positive for the positive controls even in the presence of 100 µg/mL of luspatercept.

[c] Free drug interference for ACE-536 was done in Study ACE-536-B-Thal-001 (Report 155-1809).

Source: Report 176678, Report 207-1001.

Source: EDR 2.7.1 Table 5.

4.2 Clinical PK/PD and Immunogenicity Assessments

PK Assessment

Blood samples were collected for characterization of luspatercept PK in patients with MDS, including those enrolled in Phase 2 Studies A536-03 and A536-05, as well as in Phase 3 Study ACE-536-MDS-001. Table 6 summarizes the clinical studies with the dosing regimen, drug product, and visits for PK sampling:

Table 6. Summary of clinical studies in patients with MDS.

Study (Cutoff date for report)	Dose Regimen and Drug Product	Visits for PK Sampling	No. of Subjects Included
A536-03: A Phase 2, openlabel, ascending dose study of ACE-536 for the treatment of anemia in patients with low or intermediate-1 risk myelodysplastic syndromes (MDS) (09 Aug 2017)	Dose escalation cohorts: 0.125, 0.25, 0.5, 0.75, 1, 1.33, and 1.75 mg/kg, SC, Q3W Expansion cohorts 1 & 2: Starting dose = 1 mg/kg, SC, Q3W, with intra-subject dose escalation to 1.33 and 1.75 mg/kg allowed. Drug product: 25 mg frozen liquid (Process I/II drug substance) for dose escalation cohorts and expansion cohort 1; 50 mg lyophilized powder (Process II drug substance) for expansion cohort 2.	C1D1, C1D8, C1D11, C1D15, C2D1, C2D8, C4D1, C5D1, C5D8, C5D15, EOT, post treatment, and EOS.	107
A536-05: An open-label extension study to evaluate the long-term effects of ACE-536 for the treatment of anemia in patients with low or intermediate-1 risk myelodysplastic syndromes (MDS) previously enrolled in Study A536-03 (13 Oct 2017)	Subjects without treatment interruption: Starting dose was the same as their last dose in Study A536-03, with intra-subject dose escalation to 1.33 and 1.75 mg/kg allowed Subjects with treatment interruption: Starting dose = 1 mg/kg, SC, Q3W, with intra-subject dose escalation to 1.33 and 1.75 mg/kg allowed. Drug product: 50 mg lyophilized powder for subjects who used the same drug product in Study A536-03. Switched to 50 mg lyophilized powder on a site-by-site basis for subjects who used the frozen liquid in Study A536-03.	Once every 4 cycles (C1D1, C5D1, C9D1, C13D1, C17D1, etc.) and EOS.	70
ACE-536-MDS-001: A Phase 3, double-blind, randomized study to compare the efficacy and safety of luspatercept (ACE-536) versus placebo for the treatment of anemia due to IPSS-R very low, low, or intermediate risk myelodysplastic syndromes in subjects with ring sideroblasts who require red blood cell transfusions (8 May 2018)	Starting dose = 1 mg/kg, SC, Q3W, with intra-subject dose escalation to 1.33 and 1.75 mg/kg allowed. Drug product: 25 and 75 mg lyophilized powder (Process III drug substance).	On treatment: C1D1, C1D8, C1D15, C2D1, C4D1, C5D8, C6D1, C8D1, and Week 25; then, once every 4 cycles. Posttreatment: EOT and then once every 12 weeks. Maximum 1 year of sampling from the first dose in the Primary Treatment Phase.	Active: 153 Placebo: 76

C = cycle; D = day; EOS = end of study; EOT = end of treatment; IPSS-R = International Prognostic Scoring System-Revised; MDS = myelodysplastic syndromes; No. = number; PK = pharmacokinetics; Q3W = once every three weeks; SC = subcutaneous injection.

Source: EDR 5.3.3.5 ACE-536-MPK-002 CSR Table 2.

In Study A536-03, noncompartmental PK analysis was conducted to describe individual luspatercept serum concentration-time profiles following the first dose (

Table 7). Results show that increase in mean C_{max} and AUC from time zero to 21 days (AUC_{0-21d}) was approximately proportional to dose from 0.125 to 1.75 mg/kg, and C_{max} was observed at approximately 7 days. Moreover, a preliminary one-compartment PK model was utilized with first-order absorption and elimination to describe the individual luspatercept serum concentration-time profiles upon multiple dosing for all dose levels (

Table 8). Results show that steady state was reached after 3 doses. Increases of both AUC at steady state (AUC_{ss}) and $C_{max,ss}$ were approximately proportional to dose from 0.125 to 1.75 mg/kg. The interindividual variability (IIV) of AUC_{ss} was approximately 40% based on data from the expansion cohort (N = 49 for lyophilized powder formulation).

Table 7. Summary of noncompartmental PK parameters following first dose of luspatercept in Study A536-03.

Starting Dose (mg/kg)	N	C_{max} (µg/mL)	T_{max} (day)	AUC_{0-21d} (day•µg/mL)
0.125	3	0.64 (34.9)	10 (7-14)	9.29 (32.2)
0.25	3	0.96 (93.0)	7 (6-9)	12.6 (95.0)
0.5	3	2.33 (27.2)	10 (7-15)	36.9 (4.2)
0.75	6	3.76 (42.3)	7 (7-8)	51.8 (35.9)
1.0	3	4.35 (12.9)	7 (6-9)	62.5 (25.2)
1.33	6	7.46 (14.6)	8 (6-14)	113 (17.0)
1.75	3	9.66 (7.52)	7 (6-7)	138 (1.1)
Expansion 1 [a]	31	5.80 (26.2)	7 (5-10)	78.8 (25.8)
Expansion 2 [a]	49	5.86 (24.8)	7 (6-21)	83.5 (27.1)

AUC = area under the concentration-time curve; AUC_{0-21d} = AUC from time zero to 21 days; C_{max} = maximum concentration observed in first treatment cycle; CV = coefficient of variation; N = number of subjects; T_{max} = time to reach C_{max}.

[a] Starting dose = 1 mg/kg: frozen liquid formulation for expansion 1 and lyophilized powder formulation for expansion 2.

Median (minimum – maximum) data are presented for T_{max} and geometric mean (geometric CV%) data are presented for other parameters.

Source: Report A536-03, Table 30.

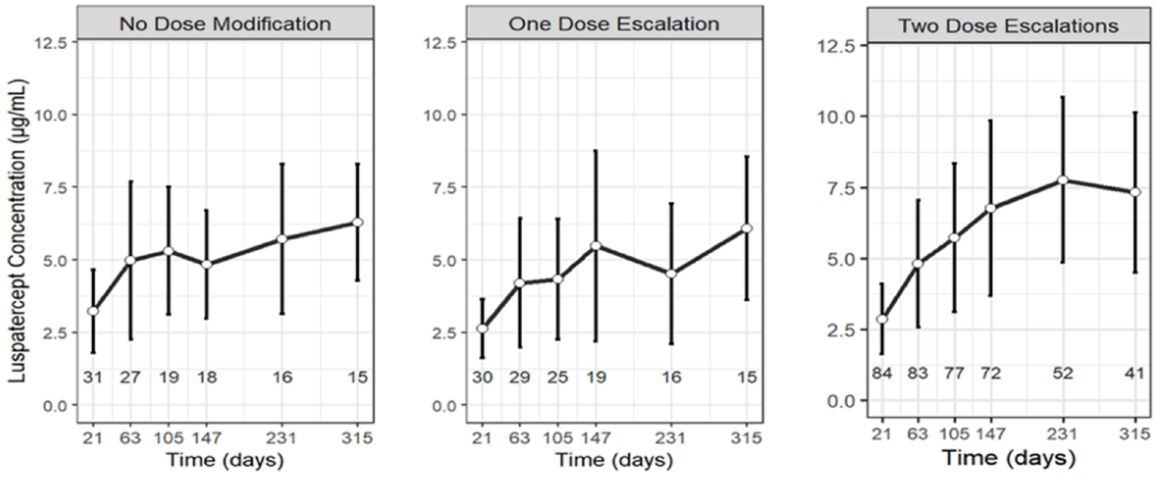
Table 8. Summary of one-compartmental PK parameters following repeated doses of luspatercept in Study A536-03.

Starting Dose (mg/kg)	N	K_a (1/day)	C_{max} (µg/mL)	T_{max} (day)	AUC_{ss} (day•µg/mL)	$t_{1/2}$ (day)	CL/F (L/day)	V1/F (L)
0.125	3	0.16 (93.7)	0.66 (30.0)	9 (8-15)	24.8 (60.9)	14.6 (48.5)	0.340 (46.5)	7.1 (36.8)
0.25	3	0.40 (126)	0.94 (74.3)	6 (3-7)	26.1 (57.5)	12.2 (113)	0.744 (82.7)	13.1 (189)
0.5	3	0.50 (250)	2.51 (31.1)	6 (2-10)	72.8 (39.8)	14.0 (44.2)	0.455 (48.2)	9.2 (38.5)
0.75	6	1.09 (117)	4.67 (49.4)	2 (1-8)	117 (37.2)	14.7 (32.5)	0.485 (36.3)	10.3 (21.8)
1.0	3	0.46 (266)	4.80 (15.9)	5 (2-9)	103 (29.2)	8.96 (48.4)	0.822 (30.9)	10.6 (50.7)
1.33	6	0.36 (59.8)	8.42 (20.5)	5 (4-12)	240 (43.1)	14.2 (38.5)	0.430 (41.2)	8.8 (24.0)
1.75	3	0.41 (251)	10.4 (18.6)	6 (2-10)	236 (10.8)	9.74 (26.4)	0.594 (9.9)	8.4 (16.2)
Expansion 1 [a]	31	0.49 (121)	6.24 (27.6)	6 (2-10)	149 (30.6)	11.5 (40.1)	0.509 (31.6)	8.4 (35.9)
Expansion 2 [a]	49	0.57 (125)	6.38 (27.8)	5 (2-11)	171 (40.0)	13.5 (45.2)	0.449 (43.5)	8.8 (26.9)

AUC_{ss} = area under the concentration-time curve at steady state for the starting dose; C_{max} = maximum concentration for the starting dose; CL/F = apparent clearance; CV = coefficient of variation; K_a = absorption rate constant; N = number of subjects; $t_{1/2}$ = elimination half-life; T_{max} = time to reach C_{max}; V1/F = apparent volume of distribution of the central compartment.
[a] Starting dose = 1 mg/kg: frozen liquid formulation for expansion 1 and lyophilized powder formulation for expansion 2.
Median (minimum - maximum) data are presented for T_{max} and geometric mean (geometric CV%) data are presented for other parameters.
Source: Report A536-03, Table 31.

The overall PK characteristics of luspatercept was assessed by population PK methodology with data from Study ACE-536-MDS-001 in combination with data from Study A536-03. In Study ACE-536-MDS-001, observed data show that in patients remaining on 1 mg/kg, mean C_{trough} was stable from Day 42 to more than 300 days; in patients with dose escalation to 1.75 mg/kg, mean C_{trough} increased by approximately 20% at later times (Day > 231) compared with patients who had no dose modifications (Figure 2). The model-predicted results in this Phase 3 study were consistent with those observed in Study A536-04, with median T_{max} as approximately 5.5 days and mean $t_{1/2}$ in serum as 11 days. The individual variability in overall exposure (AUC_{ss}) was 36%. See section below for summary of population PK report.

Figure 2. Mean trough serum concentration for luspatercept versus time in ACE-536-MDS-001.



Numbers at the bottom of each panel indicate the number of subjects at each time point.
Source: Report ACE-536-MPK-002, Figure 36.

Table 9. Summary of luspatecept PK parameters by Bayesian estimation in Study ACE-536-MDS-001.

Parameter	Responders (N = 69) [a]	Non-Responders (N = 84)	Total (N = 153)
CL/F (L/day)	0.469 (37.1)	0.559 (42.7)	0.516 (41.2)
V1/F (L)	9.11 (25.0)	10.2 (26.6)	9.68 (26.5)
$t_{1/2}$ (day)	13.5 (28.1)	12.6 (34.2)	13.0 (31.6)
T_{max} (day)	5.51 (3.88-6.69)	5.37 (3.12-6.55)	5.40 (3.12-6.69)
C_{max} (µg/mL)	6.12 (17.1)	5.50 (21.8)	5.77 (20.5)
$C_{max,ss}$ (µg/mL)	9.88 (26.6)	8.62 (31.1)	9.17 (29.9)
AUC_{ss} (day•µg/mL)	158 (33.5)	135 (40.6)	145 (38.3)

AUC = area under the concentration-time curve; AUC_{ss} = AUC at steady state for the starting dose; C_{max} = maximum concentration for the first dose; $C_{max,ss}$ = C_{max} at steady state for the starting dose; CL/F = apparent clearance; CV = coefficient of variation; N = number of subjects; $t_{1/2}$ = elimination half-life; T_{max} = time to reach C_{max}; V1/F = apparent volume of distribution of the central compartment.

[a] Responders are defined as subjects who achieved red blood cell transfusion independence ≥ 8 consecutive weeks during the first 48-week treatment period.

Median (minimum – maximum) data are presented for T_{max}; geometric mean (geometric CV%) data are presented for other parameters.

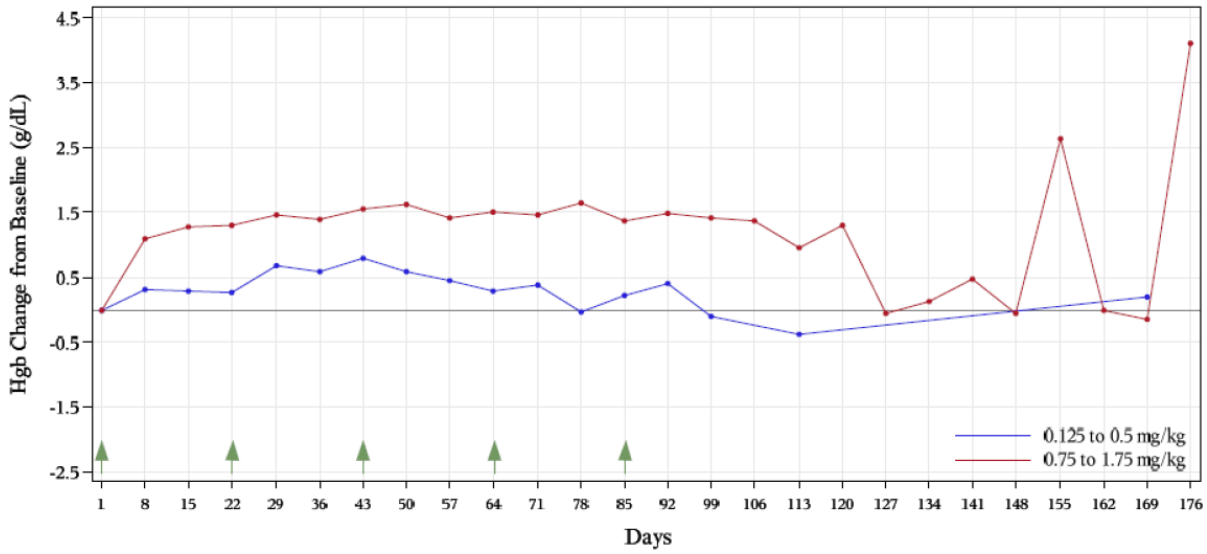
Source: Report ACE-536-MPK-002, Table 17.

PD (Erythroid Response) Assessment

Blood samples were collected prior to dosing of luspatercept to obtain the hemoglobin level. For any RBC transfusions received during the study, collect hemoglobin value just prior to transfusion.

In Study A536-03, change of hemoglobin level from baseline was assessed for patients with low transfusion burden (LTB), i.e., baseline RBC-T < 4 units/8 weeks, while RBC-T reduction was directly used to assess the erythroid response in patients with high transfusion burden (HTB), i.e., baseline RBC-T ≥ 4 units/8 weeks. In LTB patients, dose-dependent increase from baseline in Hgb was observed. The mean increase was consistently higher in the 0.75 to 1.75 mg/kg group than in the 0.125 to 0.5 mg/kg group for the duration of the study (Figure 3). The increase in Hgb was sustained through end of treatment with the Q3W dosing schedule. See Table 10 for erythroid response during a consecutive 8-week interval for the study.

Figure 3. Mean change from baseline in hemoglobin in LTB patients in Study A536-03.



Hgb = hemoglobin
Note: Baseline is defined as mean of pretreatment Hgb values between Day -28 and Day 1. Hemoglobin values within 7 days of a transfusion are excluded from the summary. Arrows show the dosing day. The sample size is ≤ 2 for the 0.125 to 0.5 mg/kg and up to 58 for the 0.75 to 1.75 mg/kg. The large fluctuation in Hgb on Days 155 and 176 in the 0.75 to 1.75 mg/kg group is due to small sample size (N = 1).
Source: Report A536-03, Figure 2.

Table 10. Erythroid response during any consecutive 8-week interval by luspatercept dose group in Study A536-03.

Baseline RBC-T Burden		**0.125 to 0.75 mg/kg**	**1 to 1.75 mg/kg**
< 4 units/8 weeks	N	5	55
	Mean Hgb increase ≥ 1.5 g/dL, n (%)	1 (20.0)	34 (61.8)
≥ 4 units/8 weeks	N	10	37
	RBC-T reduction ≥ 4 units, n (%)	3 (30.0)	20 (54.0)

Hgb = hemoglobin; RBC-T = red blood cell transfusion; N = number of subjects per treatment group; n = number of responders.
Source: Report A536-03, Table 16 and Table 17.

Immunogenicity Assessment

Blood samples for assessment of ADA in serum were collected from all subjects in all clinical studies at the following visit timepoints. Time-matched PK samples were collected to assist in the interpretation of ADA results.

- Study A536-04: Pre-dose on C1D1 and C4D1; EOT, and EOS. Additional follow-up if applicable.
- Study A536-06: Pre-dose once every 4 cycles (C1D1, C5D1, C9D1, C13D1, C17D1, etc.), EOT, and EOS. Additional follow-up if applicable.
- Study ACE-536-B-THAL-001: Pre-dose on C1D1, C2D1, C4D1, C6D1, C8D1, Week 25, once every 4 cycles (doses) thereafter and EOT. At posttreatment, collect samples on Weeks 6, 12, 24, 36, and 48 if applicable with up to 1 years of sampling.

A total of 336 patients with MDS provided evaluable ADA samples, including 260 luspatercept-treated patients and 76 placebo-treated patients. Results show that for patients in Phase 2 studies A536-03/05, 11.2% developed binding antibodies and 4.7% developed neutralized antibodies. For patients receiving lusparecept in Phase 3 study ACE-536-MDS-001, 7.2% developed binding antibodies and 3.3% developed neutralized antibody. The incidence of neutralized antibody was similar between luspatercept- and placebo-treated patients. See Table 11 for details.

Table 11. Incidence of TEADA in patients with MDS.

	Luspatercept, n (%)			Placebo, n (%)
Anti-Drug Antibodies	**A536-03/05 (N = 107)**	**ACE-536-MDS-001 (N = 153)**	**Total (N = 260)**	**ACE-536-MDS-001 (N = 76)**
ADA against luspatercept	12 (11.2)	11 (7.19)	23 (8.85)	3 (3.95)
Specificity for ACE-mECD	2 (1.87)	5 (3.27)	7 (2.69)	1 (1.32)
Specificity for ActRIIB ECD	2 (1.87)	3 (1.96)	5 (1.92)	1 (1.32)
Neutralizing luspatercept	4 (3.74)	5 (3.27)	9 (3.46)	2 (2.63)
Neutralizing ActRIIB-ECD-Fc	1 (0.93)	0 (0)	1 (0.38)	0 (0)

ActRIIB = activin receptor type IIB; ACE-mECD: modified ECD of human ActRIIB on luspatercept; ActRIIB ECD = natural ECD of human ActRIIB; ActRIIB-ECD-Fc = a fusion protein joining the natural ECD of human ActRIIB to Fc portion of human Ig G; ADA = anti-drug antibodies; ECD = extracellular domain; N = total number of subjects providing evaluable ADA sample; n = number of subjects with treatment-emergent ADA.
Source: Report ACE-536-MPK-004, Table 9.

Luspatercept dose-normalized trough concentration was analyzed and compared for patients with ADA negative, pre-existing ADA, and NAb-negative TEADA and NAb positive. Results in

Table 12 show that C_{trough} tended to be lower in patients with TEADAs (3.23 µg/mL) compared to ADA negative (4.11 µg/mL). A 37% reduction in mean C_{trough} was observed in patients with neutralizing TEADA (2.59 µg/mL) compared to ADA negative. There was no marked difference in efficacy (Table 13) and safety (Table 14) between patients with TEADA and ADA negative. However, given the percentage of patients developed NAb was small and comparable to placebo arm, no statistically meaningful comparison could be conducted to draw a conclusion.

Table 12. Summary of dose-normalized luspatercept trough concentration in serum by ADA status.

			Treatment-Emergent		
Dose-Normalized Trough Concentration (µg/mL)	**Negative (N = 220)**	**Preexisting (N = 15)**	**NAb Negative (N = 14)**	**NAb Positive (N = 9)**	**Total (N = 23)**
Mean (CV%)	4.11 (45.7)	3.81 (32.6)	3.64 (39.6)	2.59 (64.3) [a]	3.23 (49.0)
90% CI of mean	3.90-4.32	3.24-4.37	2.96-4.32	1.56-3.62	2.66-3.80

ADA = anti-drug antibodies; CI = confidence interval; CV = coefficient of variation; N = number of subjects; NAb = neutralizing anti-drug antibody against luspatercept.
[a] p = 0.018 versus ADA negative subjects (one-way analysis of variance).
Source: Report ACE-536-MPK-004, Table 13.

Table 13. Summary of selected efficacy endpoints by ADA status.

Efficacy Endpoint	Negative		Preexisting		Treatment-Emergent	
	N	n (%)	N	n (%)	N	n (%)
RBC-TI ≥ 8 weeks (Weeks 1-15) [a]	184	63 (34.2)	12	7 (58.3)	18	5 (27.8)
RBC-TI ≥ 8 weeks (Weeks 1-24) [b]	135	49 (36.3)	7	4 (57.1)	11	5 (45.5)
RBC-TI ≥ 12 weeks (Weeks 1-24) [b]	135	38 (28.1)	7	3 (42.9)	11	2 (18.2)

N = number of subjects; n = number of responders; RBC-T = red blood cell transfusion; RBC-TI = red blood cell transfusion independency.

[a] Based on data pooled from Study A536-03 and Study ACE-536-MDS-001. Only subjects who received a starting dose ≥ 1 mg/kg and had a baseline RBC-T burden ≥ 2 units/8 weeks are included.

[b] Based on data from Study ACE-536-MDS-001 only.

Source: Report ACE-536-MPK-004, Table 15.

Table 14. Incidence of Immunogenicity Like Reactions by ADA status.

	Anti-Drug Antibody Status					
	Negative		Preexisting		Treatment-Emergent	
Treatment	N	n (%)	N	n (%)	N	n (%)
Luspatercept	222	19 (8.56)	15	2 (13.3)	23	3 (13.0)
Placebo	71	4 (5.63)	2	0 (0)	3	0 (0)

N = total number of subjects; n = number of subjecs with the defined TEAE.

Cutoff date is 08 May 2018 for Study ACE-536-MDS-001 and up to 48 weeks from the first dose for Studies A536-03 and A536-05.

Source: Report ACE-536-MPK-004, Table 16.

4.3 Population PK Analysis

Population PK analysis was conducted using 2403 evaluable luspatercept concentrations in 260 patients from Trials A536-03 and ACE-536-MDS-001. Summary statistics of the continuous and categorical covariates that were evaluated in the population PK analysis are shown in *Table 15* and *Table 16*, respectively.

Table 15. Summary Statistics for the Continuous Covariates in the Population PK Analysis

Characteristics	A536-03 N = 107		ACE-536-MDS-001 N = 153		Total N = 260	
	Mean (CV%)	Median [Min, Max]	Mean (CV%)	Median [Min, Max]	Mean (CV%)	Median [Min, Max]
Age (years)	70.6 (15.3)	72.0 [27.0, 90.0]	70.5 (12.3)	71.0 [40.0, 95.0]	70.5 (13.6)	72.0 [27.0, 95.0]
Weight (kg)	77.3 (18.5)	77.0 [48.0, 110]	76.2 (19.8)	76.0 [46.0, 124]	76.6 (19.2)	76.3 [46.0, 124]
Erythropoietin (U/L)[a]	350 (126.5)	160 [9.80, 2030]	219 (148.8)	117 [10.4, 2450]	273 (140.4)	138 [9.80, 2450]
Transfusion Burden (units/24 weeks)	11.6 (83.6)	11.5 [0.00, 40.2]	18.3 (44.8)	16.6 [5.46, 43.4]	15.5 (60.6)	15.1 [0.00, 43.4]
Total Bilirubin (µmol/L)	15.0 (60.7)	12.3 [4.62, 52.2]	17.7 (57.0)	15.0 [4.00, 68.0]	16.6 (58.9)	14.0 [4.00, 68.0]
Albumin (g/L)	43.4 (10.1)	44.0 [31.0, 52.6]	44.2 (7.6)	44.0 [35.0, 51.0]	43.9 (8.7)	44.0 [31.0, 52.6]
Alkaline Phosphatase (U/L)	80.5 (55.6)	73.0 [36.0, 301]	73.6 (32.9)	69.0 [26.0, 156]	76.4 (44.8)	70.0 [26.0, 301]
Alanine Transaminase (U/L)	34.5 (81.5)	25.0 [5.00, 190]	32.1 (75.8)	23.0 [6.00, 133]	33.1 (78.4)	23.5 [5.00, 190]
Aspartate Transaminase (U/L)	28.3 (60.0)	22.8 [8.00, 96.0]	23.9 (57.1)	19.0 [7.00, 77.0]	25.7 (59.3)	21.0 [7.00, 96.0]
Lactate Dehydrogenase (U/L)	214 (31.4)	203 [107, 620]	182 (35.1)	168 [96.0, 434]	195 (34.3)	185 [96.0, 620]
Estimated Glomerular Filtration Rate (mL/min/1.73 m^2)	72.8 (28.6)	69.6 [29.6, 128]	78.4 (28.9)	77.4 [29.7, 150]	76.1 (29.0)	73.1 [29.6, 150]

Source: Clinical PK/PD Report ACE-536-MPK-002, Table 7.

Table 16. Summary Statistics for the Categorical Covariates in the Population PK Analysis

Characteristics	Number (%) of Subjects		
	A536-03 N = 107	ACE-536-MDS-001 N = 153	Total N = 260
Baseline Characteristics			
Sex			
Female	42 (39.3%)	59 (38.6%)	101 (38.8%)
Male	65 (60.7%)	94 (61.4%)	159 (61.2%)
Race			
White	107 (100%)	107 (69.9%)	214 (82.3%)
Black	0 (0%)	1 (0.7%)	1 (0.4%)
Other (mostly unreported)	0 (0%)	45 (29.4%)	45 (17.3%)
Transfusion Dependence			
Dependent (≥ 2 RBC units/8 weeks)	73 (68.2%)	153 (100%)	226 (86.9%)
Independent (< 2 RBC units/8 weeks)	34 (31.8%)	0 (0%)	34 (13.1%)
Hepatic Function Categories			
Normal	58 (54.2%)	96 (62.7%)	154 (59.2%)
Mild	38 (35.5%)	44 (28.8%)	82 (31.5%)
Moderate	11 (10.3%)	12 (7.8%)	23 (8.8%)
Severe	0 (0%)	1 (0.7%)	1 (0.4%)
Renal Function Categories			
Normal	20 (18.7%)	50 (32.7%)	70 (26.9%)
Mild	62 (57.9%)	72 (47.1%)	134 (51.5%)
Moderate	25 (23.4%)	31 (20.3%)	56 (21.5%)
Ring Sideroblasts Positive			
Yes	63 (58.9%)	153 (100%)	216 (83.1%)
No	30 (28.0%)	0 (0%)	30 (11.5%)
Unknown	14 (13.1%)	0 (0%)	14 (5.4%)
IPSS-R Risk			
Very Low	2 (1.9%)	18 (11.8%)	20 (7.7%)
Low	60 (56.1%)	109 (71.2%)	169 (65.0%)
Intermediate	35 (32.7%)	25 (16.3%)	60 (23.1%)
High	9 (8.4%)	1 (0.7%)	10 (3.8%)
Very High	1 (0.9%)	0 (0%)	1 (0.4%)

Characteristics	Number (%) of Subjects		
	A536-03 N = 107	ACE-536-MDS-001 N = 153	Total N = 260
On Treatment Characteristics			
Drug Product			
25 mg frozen liquid (Process I/II)	58 (54.2%)	0 (0%)	58 (22.3%)
50 mg lyophilized powder (Process II)	49 (45.8%)	0 (0%)	49 (18.8%)
25 or 75 mg lyophilized powder (Process III)	0 (0%)	153 (100%)	153 (58.8%)
Concurrent Use of Iron Chelation Therapy			
Yes	28 (26.2%)	72 (47.1%)	100 (38.5%)
No	79 (73.8%)	81 (52.9%)	160 (61.5%)
Antidrug Antibodies Status on Treatment			
Negative	87 (81.3%)	135 (88.2%)	222 (85.4%)
Pre-existing	8 (7.5%)	7 (4.6%)	15 (5.8%)
Treatment-emergent	12 (11.2%)	11 (7.2%)	23 (8.8%)

Source: Clinical PK/PD Report ACE-536-MPK-002, Table 6.

The PK of luspatercept was best characterized by a one-compartment model with first order absorption rate constant (ka) and linear elimination. The inter-individual variability (IIV) was estimated as log-normally distributed with a non-zero covariance on apparent clearance (CL/F), apparent volume of distribution (V/F), with a block omega on CL/F and V/F.

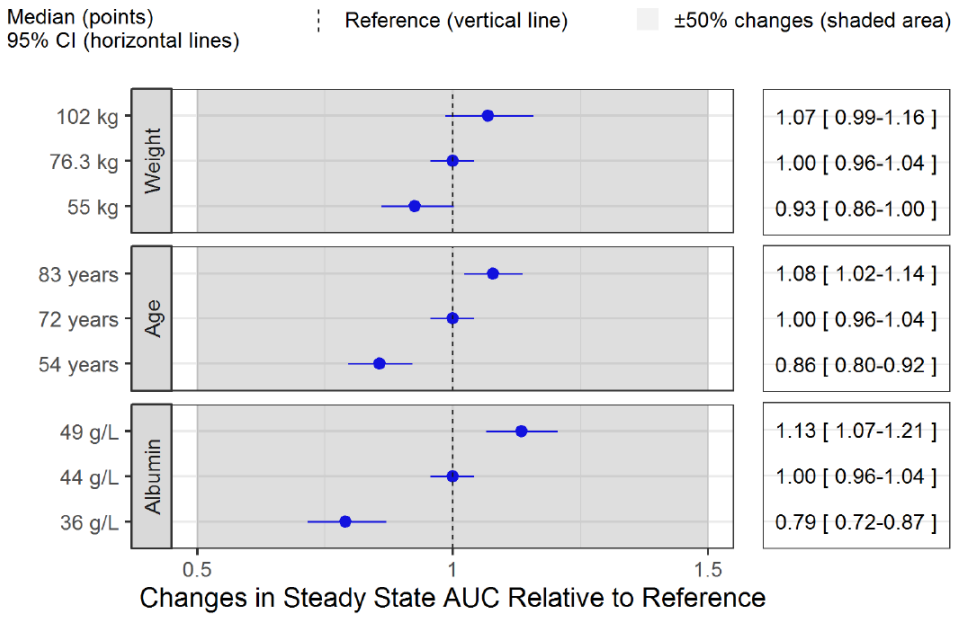
Covariate analysis identified 3 statistically significant and clinically relevant covariates, including the effects of weight, age and baseline albumin on CL/F, and the effects of weight and baseline albumin on V/F. Sex, race (Asian vs. non-Asian), mild to moderate renal impairment, mild to severe hepatic impairment defined by NCI-ODWG criteria, baseline liver enzymes (AST and ALT), baseline total bilirubin, baseline transfusion burden, baseline EPO, positive ring sideroblasts, location of SC injection (i.e., upper arm, thigh, or abdomen), drug substance manufacturing process and drug product formulation (Process I/II frozen liquid vs. Process III lyophilized powder), and concurrent iron chelation therapy had no clinically meaningful effect on luspatercept PK. The equations of the final covariate models are the following:

$$\text{CL/F (L/day)} = 0.469 \times \left(\frac{\text{Weight}}{70}\right)^{0.769} \times \left(\frac{\text{Age}}{72}\right)^{-0.534} \times \left(\frac{\text{Albumin}}{44}\right)^{-1.17}$$

$$\text{V1/F (L)} = 9.22 \times \left(\frac{\text{Weight}}{70}\right)^{0.877} \times \left(\frac{\text{Albumin}}{44}\right)^{-0.610}$$

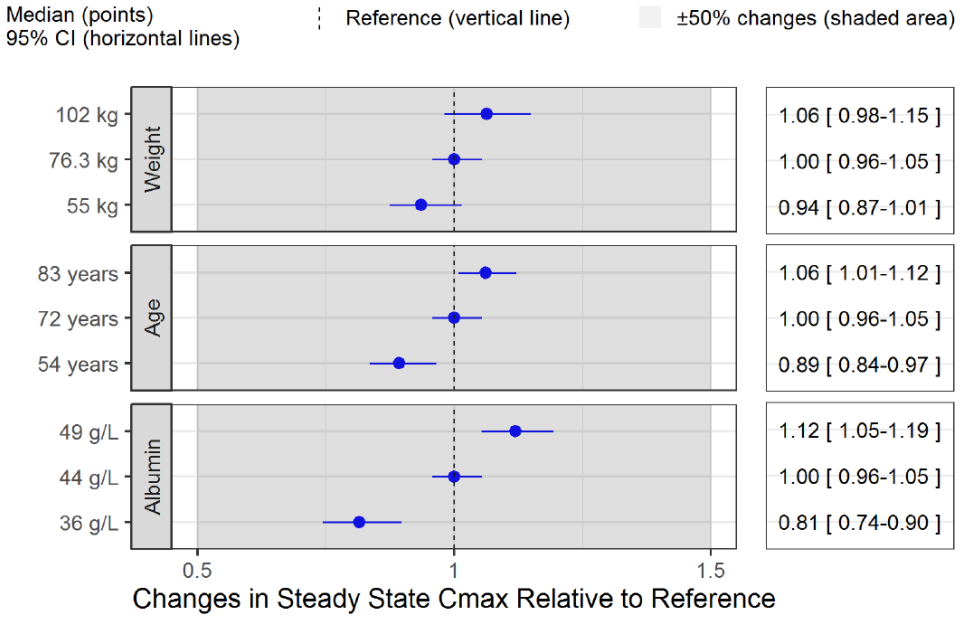
The effects of each covariate retained in the final PK model on steady-state AUC and C_{max} are presented in Figure 4 and Figure 5, respectively. At the body weight-based dose of 1 mg/kg, no clinically significant difference (< 25%) was expected in the median exposure level between the 5th or 95th quantile of each covariate and the reference value.

Figure 4. Forest Plot of Significant Covariates on Steady State AUC in the Final Model



Source: Clinical PK/PD Report ACE-536-MPK-002, Appendix A, Section 4.24.

Figure 5. Forest Plot of Significant Covariates on steady State C_{max} in the Final Model



Source: Clinical PK/PD Report ACE-536-MPK-002, Appendix A, Section 4.25.

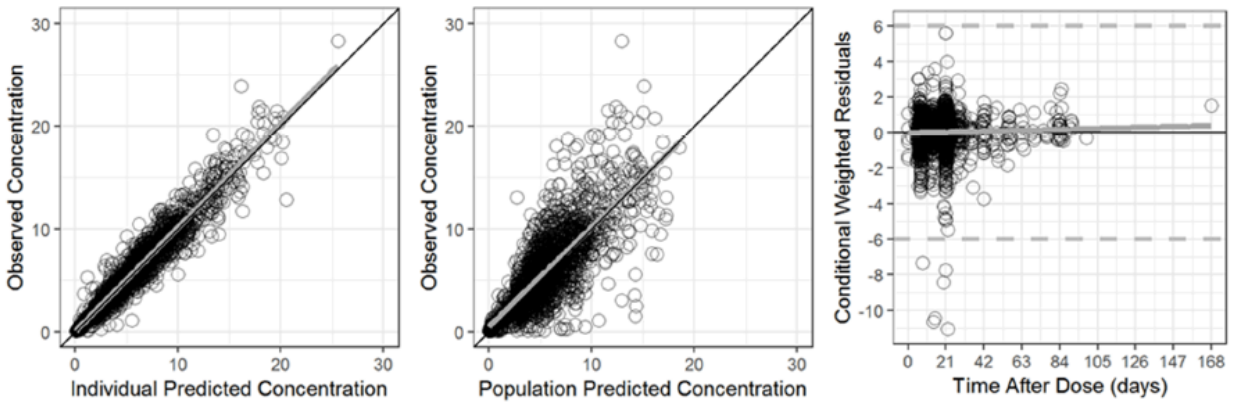
The final luspatercept PK model parameter estimates and the corresponding 95% confidence interval (CI) from Bootstrap are presented in Table 17. The goodness-of-fit plots for the final luspatercept PK model are presented Figure 6. The prediction-corrected visual predictive check (pcVPC) stratified by study (Figure 7) illustrated the prediction percentiles and corresponding 95% CI of simulated concentrations overlaid on the observed luspatercept concentrations and the corresponding 5th and 95th percentiles.

Table 17. Population PK Parameters of Luspatercept from the Final PK Model and Bootstrap

Parameter	Model Term	Estimate	Asymptotic		Bootstrap	
			RSE (%)	95% CI	Median	95% CI
CL/F (L/day)	θ	0.469	2.22	0.449 – 0.490	0.469	0.449 – 0.489
Weight (kg)	$\times (WT/70)^{\theta}$	0.769	14.2	0.555 – 0.984	0.768	0.561 – 0.986
Age (years)	$\times (AGE/72)^{\theta}$	-0.534	19.1	-0.734 – -0.334	-0.534	-0.764 – -0.315
Albumin (g/L)	$\times (ALB/44)^{\theta}$	-1.17	18.4	-1.58 – -0.746	-1.18	-1.61 – -0.726
V1/F (L)	θ	9.22	1.85	8.89 – 9.55	9.20	8.88 – 9.52
Weight (kg)	$\times (WT/70)^{\theta}$	0.877	10.2	0.702 – 1.05	0.878	0.709 – 1.05
Albumin (g/L)	$\times (ALB/44)^{\theta}$	-0.610	32.4	-0.997 – -0.223	-0.609	-1.01 – -0.216
K_a (day^{-1})	θ	0.456	11.1	0.357 – 0.554	0.456	0.383 – 0.652
Interindividual Variability						
On CL/F	$\omega = SD(\eta_{CL,i})$	0.353	7.03	0.304 – 0.401	0.349	0.304 – 0.393
On V1/F	$\omega = SD(\eta_{V1,i})$	0.222	11.8	0.171 – 0.274	0.220	0.169 – 0.271
Correlation CL/F, V1/F	$\omega = Corr(\eta_{CL,i}, \eta_{V1,i})$	0.511	13.5	0.375 – 0.647	0.516	0.363 – 0.638
Residual Variability						
Log-additive error	$\sigma = SD(\varepsilon_{i,j})$	0.221	11.2	0.173 – 0.270	0.220	0.177 – 0.270

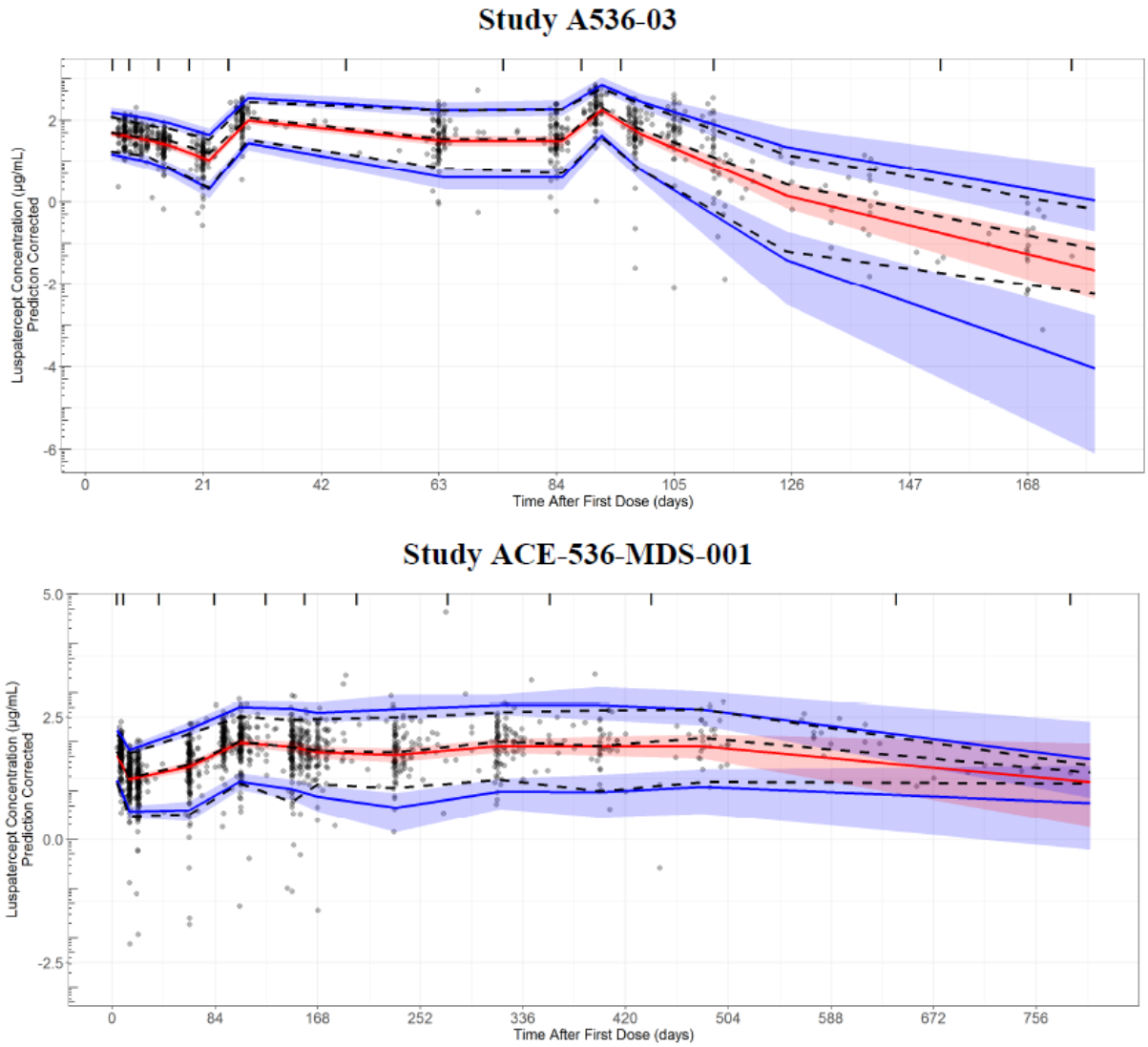
Source: Clinical PK/PD Report ACE-536-MPK-002, Table 8.

Figure 6. Goodness-of-fit Plots for the Final Population PK Model for Luspatercept



Source: Clinical PK/PD Report ACE-536-MPK-002, Figure 11.

Figure 7. Prediction Corrected Visual Predictive Check for the Final Luspatercept Model



Source: Clinical PK/PD Report ACE-536-MPK-002, Figure 12.

Reviewer's Comments: The Applicant's population PK model appears adequate to describe the luspatercept serum concentration-time profiles following the administration of luspatercept ranged from 0.125 mg/kg to 1.75 mg/kg SC every three weeks in patients with MDS. The shrinkage value was 2.5% for IIV on CL/F, 18.8% for IIV on V/F, and 8.8% for residual variability, indicating there was no obvious bias in the parameter estimates. Therefore, the PK model is acceptable for simulating post-hoc exposure metrics, e.g. average AUC from Week 1 to Week 15 (AUC_{avg15}) and average AUC to the first AE event (AUC_{avg}) of luspatercept for exposure-response analyses for efficacy and safety measurements.

Covariate analysis identified three statistically significant covariates: age, body weight, and baseline albumin on CL/F, as well as body weight and baseline albumin on Vd/F. The significant impact of body weight on luspatercept CL/F and Vd/F supported the body-weight-based dosing regimen. Simulations

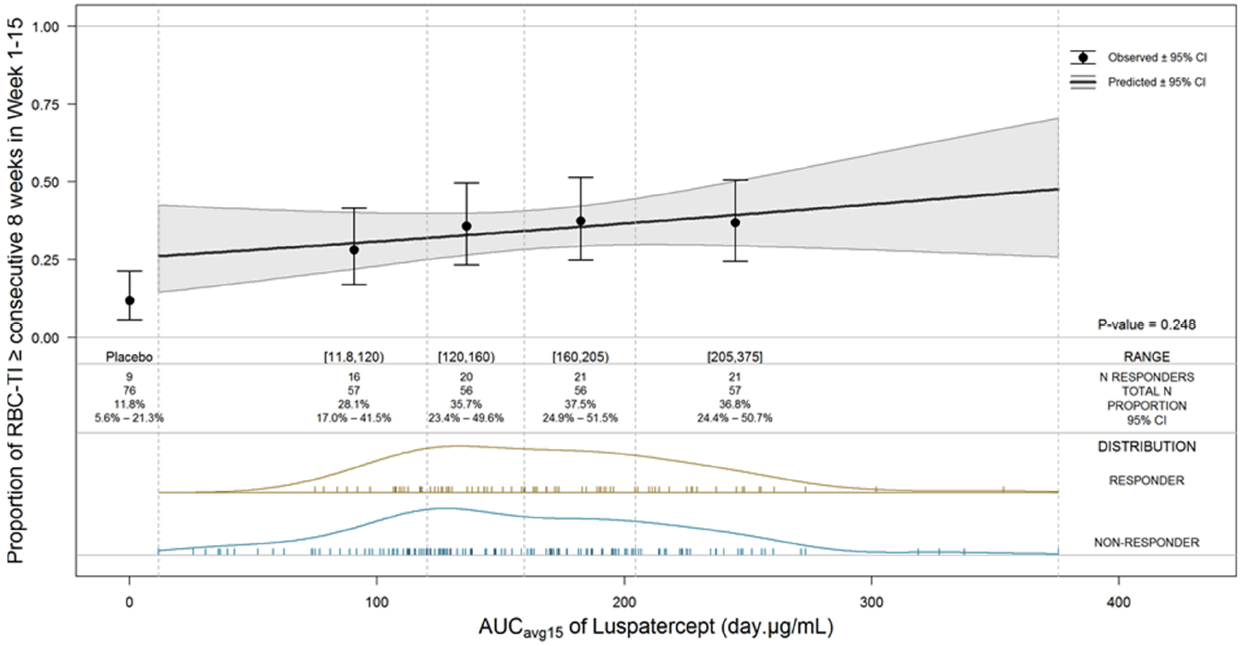
utilizing the final population PK model revealed that the impact of age and baseline albumin on luspatercept serum exposure was limited with < 20% alterations with body weight-based dosing and hence not clinically significant. In addition, the luspatercept exposure was not clinically significantly altered by sex, race, mild to severe hepatic impairment, mild to moderate renal impairment, baseline serum erythropoietin, baseline transfusion burden, positive ring sideroblasts, location of SC injection, and concurrent iron chelation therapy after the dose was adjusted by body weight. Therefore, no dose adjustment is needed for the above-mentioned specific populations.

4.4 Exposure-Response Analysis

Applicant's Exposure-Response for Efficacy

The exposure-response (E-R) analysis was conducted for proportion of patients achieving RBC transfusion independence (RBC-T) with a duration of ≥ 8 weeks measured at Week 15 in 222 patients with MDS and regular transfusions of ≥ 2 RBC units/8 weeks at baseline from Trials A536-03 and ACE-536-MDS-001. The dose of luspatercept ranged from 0.125 to 1.75 mg/kg for the pooled population. However, the proportion of patients who started with doses < 1 mg/kg was small, with only 0.9% (2/222) patients on 0.125 mg/kg, 0.9% (2/222) patients on 0.25 mg/kg, 1.4% (3/222) patients on 0.5 mg/kg, and 2.7% (6/222) patients on 0.75 mg/kg. As such, the exposure range was narrow (mainly 1 to 1.75 mg/kg) for the integrated analysis. The PK metrics used for E-R analysis was AUC_{avg15}. The Applicant concluded that a positive exposure-dependent (AUC_{avg15}) trend was observed for RBC-TI for ≥ 8 consecutive weeks during the first 15 weeks, but this apparent trend was not statistically significant in luspatercept-treated patients (Figure 8).

Figure 8. Relationship between Luspatercept Serum Exposure and Probability of Achieving RBC-TI ≥ 8 Consecutive Weeks in Week 1 to Week 15



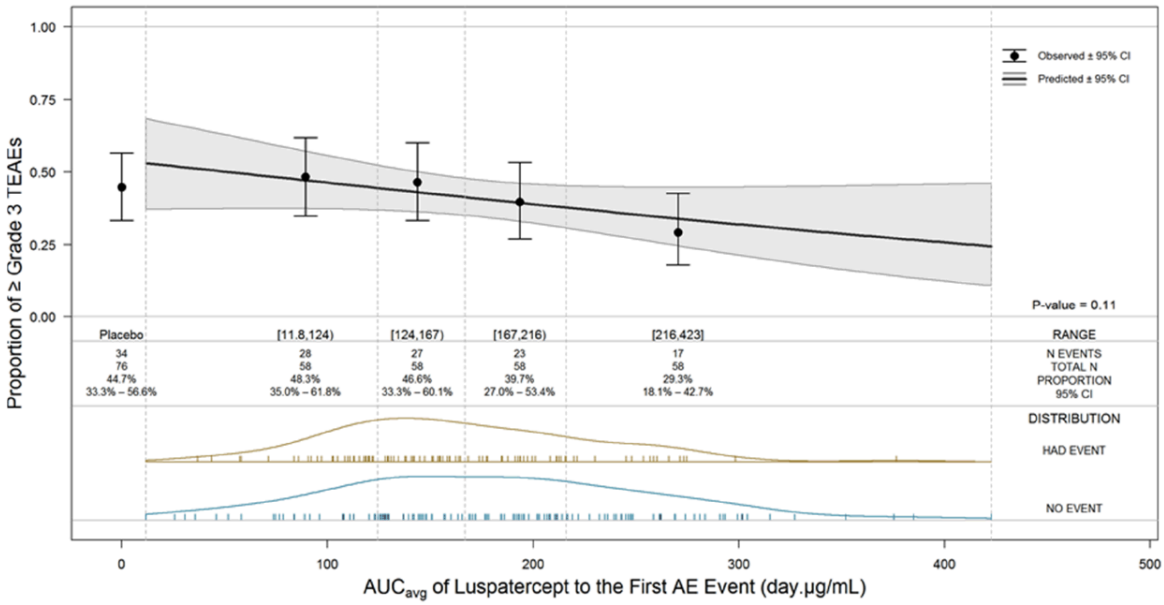
Source: Clinical PK/PD Report ACE-536-MPK-002, Figure 15.

Reviewer's Comments: The pooled exposure-efficacy analysis was confounded by dose titration design implemented in both Trials A536-03 and ACE-536-MDS-001, since the titration was based on individual patient's response. In the dose escalation part in Trial A536-03, higher response rate was observed with higher doses (0.75-1.75 mg/kg Q3W vs. 0.125 to 0.5 mg/kg Q3W).

Applicant's Exposure-Response for Safety

The exposure-safety analysis was conducted in 372 (76 placebo and 263 luspatercept) patients with MDS and regular transfusions of ≥ 2 RBC units/8 weeks at baseline from Trials A536-03, A536-05 and ACE-536-MDS-001. The dose of luspatercept ranged from 0.125 to 1.75 mg/kg for the pooled population. The PK metrics used for E-R analysis was average area under the concentration-time curve to the first event (AUC_{avg}). The Applicant concluded that there was an inverse relationship between luspatercept AUC_{avg} and Grade ≥3 TEAEs (Figure 9), but the frequency of ≥ Grade 3 TEAEs in each exposure group was statistically similar to that observed in the placebo-treated patients prior to accounting for the effect of baseline risk factors.

Figure 9. Relationship between Luspatercept Serum Exposure and Probability of Experiencing TEAEs ≥ Grade 3



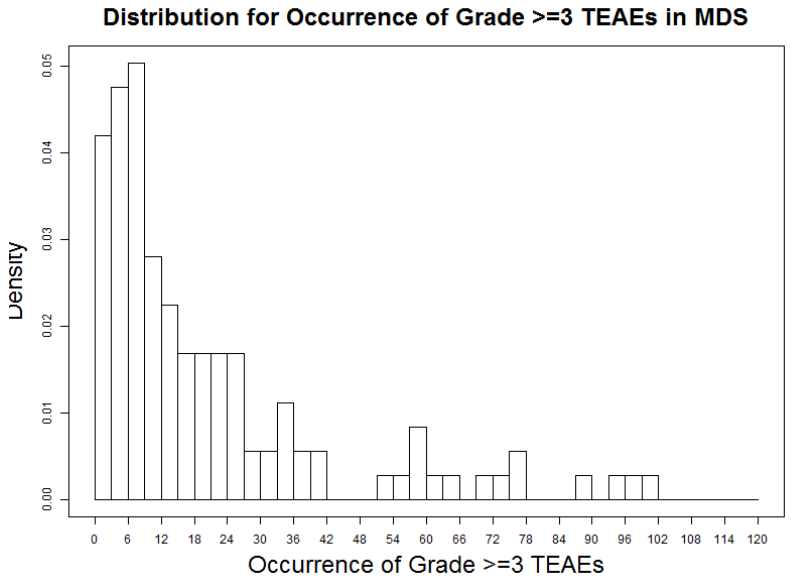
Source: Clinical PK/PD Report ACE-536-MPK-002, Figure 27.

Reviewer's Comments: The Applicant's inverse exposure-safety relationship was potentially confounded by the timing of TEAE occurrence and dose escalation scheme. The exposure-response relationship for safety was corrected by the reviewer's sensitivity analysis which confirmed that the relationship between exposure and safety is flat at Weeks 0 to 6 and positive at Week 6 and afterward.

*The reviewer noted that most Grade ≥3 TEAEs occurred in the first two treatment cycles (0-6 weeks) prior to dose escalation (*Figure 10*), where the patients were still on the starting dose and the concentrations associated with these events would be lower compared to that in the later phase. Therefore, the analyses for safety were confounded by the trial design (dose titration) and cannot represent the true E-R relationship. Accordingly, the reviewer conducted sensitivity analysis to explore the E-R for safety by different time periods. The biased relationship was then corrected and the results showed that the E-R relationships were generally flat for Grade ≥3 TEAEs before dose escalation at Weeks 0-6 (*Figure 11*) and*

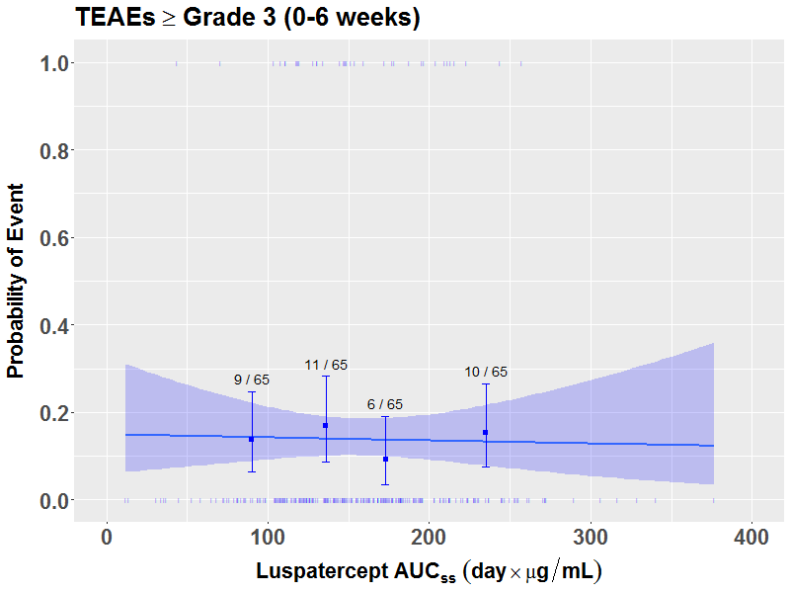
*positive during dose escalation at Week 6 and afterwards (*Figure 12*), suggesting that increase of luspatercept exposure up to 1.75 mg/kg was associated with the slight increase in the occurrence of these AEs.*

Figure 10. Distribution of occurrence of Grade ≥3 TEAEs



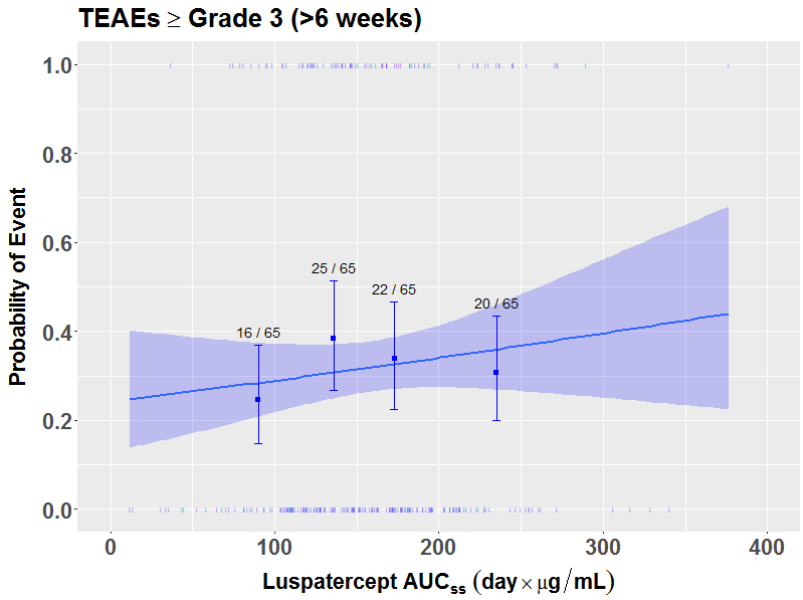
Source: Reviewer's analysis.

Figure 11. Relationship between Luspatercept Serum Exposure and Probability of Experiencing TEAEs ≥ Grade 3 before Dose Escalation at Weeks 0 to 6



Source: Reviewer's analysis.

Figure 12. Relationship between Luspatercept Serum Exposure and Probability of Experiencing TEAEs ≥ Grade 3 during Dose Escalation after Week 6.



Source: Reviewer's analysis.

4.5 Dose-Neoplasm Analysis

Based on pooled safety analysis in patients with lower-risk MDS (excluding 10 patients with high and very high risk MDS) from studies ACE-MDS-001, A536-03 and A536-05 (cutoff date of 7/1/2019), there were 46 events of MDS progression, AML and SPM in patients who received luspatercept, including 25 events from Phase 2 studies A536-03/05 and 21 events from luspatercept arm of Phase 3 study ACE-536-MDS-001, while only 5 events were observed in placebo arm of the Phase 3 study (Table 18).

Table 18. Events of MDS Progression, AML, and SPM in Studies ACE-MDS-001, A536-03 and A536-05

Event Category	Luspatercept arm in Study A536-03/05	Luspatercept arm in Study ACE-536-MDS-001	Placebo arm in Study ACE-536-MDS-001
MDS progression/AML	11	10	4
SPM	14	11	1

Source: Reviewer's analysis.

As shown in Table 19, the incidence rate of MDS progression/AML/SPM events tended to be higher in luspatercept group vs. placebo group (15% vs. 7%), especially for SPM events (8% vs. 1%). Median [range] time to MDS/AML/SPM events tended to be shorter in luspatercept group (383 [36, 1295] days) vs. placebo group (624 [162, 807] days). However, there is no clear association between the incidence rates of those events and the maximum dose level patients received. Further analysis suggests that the incidence rates are not clearly associated with drug exposure, baseline EPO or RBC-T burden.

Table 19. Dose-Neoplasm Analysis based on Pooled Safety Data in Patients with Lower-Risk MDS from Studies ACE-MDS-001, A536-03 and A536-05

Parameter	Placebo (n=76)	Luspatercept Maximum Dose Level			All luspatercept (n=259)
		⩽1 mg/kg (n=45)	1.33 mg/kg (n=38)	1.75 mg/kg (n=176)	
No. of MDS/AML/SPM events	5	9	12	25	46
MDS progression/AML events	4	4	8	9	21
SPM events	1	5	4	16	25
No. (%) of pts with MDS/AML/SPM events	5 (7%)	7 (16%)	12 (32%)	21 (12%)	40 (15%)
MDS progression/AML events	4 (5%)	4 (9%)	8 (21%)	9 (5%)	21 (8%)
SPM events	1 (1%)	4 (9%)	4 (11%)	13 (7%)	21 (8%)
Time to MDS/AML/SPM events (days)	624 [162, 807]	381 [36, 1035]	422 [50, 1193]	597 [101, 1295]	383 [36, 1295]
Treatment duration (days)					
pts with MDS/AML/SPM events	148 [106, 150]	342 [1, 1494]	453 [43, 1494]	462 [106, 1438]	462 [1, 1494]
pts without MDS/AML/SPM events	148 [51, 701]	130 [1, 1184]	193 [49, 1029]	305 [43, 1528]	211 [1, 1528]
Geomean (CV%) AUC_{AVG48} **(µg*day/mL)**					
pts with MDS/AML/SPM events		121 (58%)	144 (30%)	192 (39%)	165 (44%)

pts without MDS/AML/SPM events		113 (79%)	146 (40%)	177 (51%)	159 (58%)
Median [range] baseline EPO (U/L)					
pts with MDS/AML/SPM events	626 [54, 1223]	83 [10, 201]	98 [26, 542]	90 [12, 1644]	88 [10, 1644]
pts without MDS/AML/SPM events	124 [29, 2760]	159 [30, 1268]	163 [15, 2433]	154 [0, 2454]	160 [0, 2454]
Median [range] baseline RBC-T					
Trial MDS-001 (units/16 week)					
pts with MDS/AML/SPM events	13 [4, 18]	6 [6, 6]	15 [8, 25]	9 [5, 18]	8 [5, 25]
pts without MDS/AML/SPM events	10 [4, 40]	10 [2, 27]	12 [4, 24]	10 [4, 30]	10 [2, 30]
Trials 03/05 (units/8 week)					
pts with MDS/AML/SPM events		2 [2, 4]	5 [4, 6]	5 [2, 14]	4 [2, 14]
pts without MDS/AML/SPM events		4 [2, 6]	4 [2, 8]	4 [2, 18]	4 [2, 18]

Source: Reviewer's analysis.

Similar analysis was performed by study as shown in Table 20 for study ACE-MDS-001 and Table 21 for studies A536-03 and A536-05. Patients in studies A536-03/05 tended to have a higher risk of disease progression and/or SPM compared to patients in luspatercept arm of study ACE-MDS-001. The Sponsor argued that studies A536-03/05 included a broader and more heterogeneous population of patients than did the study ACE-MDS-001. All patients enrolled in study ACE-MDS-001 were ring sideroblast (RS)-positive, while 62.2% (66/106) patients with lower-risk MDS enrolled in studies A536-03/05 were RS-positive. The Applicant found that 9 of the 12 patients who progressed to high-risk MDS or AML in studies A536-03/05 were non-RS patients, and 8 harbored adverse molecular mutations at baseline including ASXL1 or TP53. The Applicant believed that both the non-RS phenotype and the adverse mutational profile impart a poorer prognosis and a more rapid progression to higher risk disease and AML, irrespective of IPSS-R risk score.

Table 20. Dose-Neoplasm Analysis based on Safety Data in Patients with Lower-Risk MDS from Study ACE-MDS-001.

Parameter		Luspatercept Maximum Dose Level			All luspatercept (n=153)
	Placebo (n=76)	≤1 mg/kg (n=25)	1.33 mg/kg (n=25)	1.75 mg/kg (n=103)	
No. of MDS/AML/SPM events	5	3	4	14	21
MDS progression/AML events	4	2	4	4	10
SPM events	1	1	0	10	11
No (%) of pts with MDS/AML/SPM events	5 (7%)	2 (8%)	4 (16%)	11 (11%)	17 (11%)
MDS progression/AML events	4 (5%)	2 (8%)	4 (16%)	4 (4%)	10 (7%)
SPM events	1 (1%)	1 (4%)	0 (0%)	7 (7%)	8 (5%)
Time to MDS/AML/SPM events (days)	624 [162, 807]	378 [147, 690]	475 [50, 710]	380 [104, 918]	379 [50, 918]
Treatment duration (days)					
pts with MDS/AML/SPM events	148 [106, 150]	322 [322, 659]	450 [43, 722]	306 [127, 918]	322 [43, 918]
pts without MDS/AML/SPM events	148 [51, 701]	148 [21, 1184]	197 [49, 1029]	429 [85, 1079]	364 [21, 1184]

Geomean (CV%) AUC_{AVG48} **(µg*day/mL)**					
pts with MDS/AML/SPM events		89 (82%)	130 (7%)	183 (39%)	155 (50%)
pts without MDS/AML/SPM events		137 (37%)	147 (29%)	191 (43%)	173 (43%)
Median [range] baseline EPO (U/L)					
pts with MDS/AML/SPM events	626 [54, 1223]	201 [152, 201]	222 [32, 542]	117 [12, 1076]	152 [12, 1076]
pts without MDS/AML/SPM events	124 [29, 2760]	145 [30, 1184]	161 [24, 514]	164 [19, 2454]	158 [19, 2454]
Median [range] baseline RBC-T (units/16 week)					
pts with MDS/AML/SPM events	13 [4, 18]	6 [6, 6]	15 [8, 25]	9 [5, 18]	8 [5, 25]
pts without MDS/AML/SPM events	10 [4, 40]	10 [2, 27]	12 [4, 24]	10 [4, 30]	10 [2, 30]

Source: Reviewer's analysis.

Table 21. Dose-Neoplasm Analysis based on Pooled Safety Data in Patients with Lower-Risk MDS from Studies A536-03 and A536-05

Parameter	Luspatercept Maximum Dose Level			All luspatercept (n=106)
	≤1 mg/kg (n=11)	1.33 mg/kg (n=25)	1.75 mg/kg (n=70)	
No. of MDS/AML/SPM events	6	8	11	25
MDS progression/AML	2	4	5	11
SPM events	4	4	6	14
No (%) of pts with MDS/AML/SPM events	5 (45%)	8 (32%)	10 (14%)	23 (22%)
MDS progression/AML	2 (18%)	4 (16%)	5 (7%)	11 (10%)
SPM events	3 (27%)	4 (16%)	6 (9%)	13 (12%)
Time to MDS/AML/SPM events (days)	383 [36, 1035]	422 [64, 1193]	708 [101, 1295]	463 [36, 1295]
Treatment duration (days)				
pts with MDS/AML/SPM events	483 [1, 1494]	453 [44, 1494]	617 [106, 1438]	526 [1, 1494]
pts without MDS/AML/SPM events	86 [85, 573]	85 [1, 1108]	86 [42, 1528]	86 [1, 1528]
Geomean (CV%) AUC_{AVG48} **(µg*day/mL)**				
pts with MDS/AML/SPM events	153 (27%)	151 (37%)	205 (39%)	175 (39%)
pts without MDS/AML/SPM events	70 (142%)	147 (52%)	161 (57%)	143 (73%)
Median [range] baseline EPO (U/L)				
pts with MDS/AML/SPM events	45 [10, 98]	98 [26, 326]	90 [42, 1644]	83 [10, 1644]
pts without MDS/AML/SPM events	113 [47, 1268]	269 [15, 2433]	146 [0, 2032]	156 [0, 2433]
Median [range] baseline RBC-T (units/8 week)				
pts with MDS/AML/SPM events	2 [2, 4]	5 [4, 6]	5 [2, 14]	4 [2, 14]
pts without MDS/AML/SPM events	4 [2, 6]	4 [2, 8]	4 [2, 18]	4 [2, 18]

Source: Reviewer's analysis.

This is a representation of an electronic record that was signed electronically. Following this are manifestations of any and all electronic signatures for this electronic record.

/s/

LILI PAN
12/20/2019 04:39:38 PM

LIANG LI
12/20/2019 04:49:41 PM

LIAN MA
12/20/2019 05:29:11 PM

GUOXIANG SHEN
12/20/2019 05:39:39 PM

BRIAN P BOOTH
12/23/2019 11:13:47 AM