# CENTER FOR DRUG EVALUATION AND RESEARCH

***APPLICATION NUMBER:***

# 761046Orig1s000

# CLINICAL PHARMACOLOGY AND BIOPHARMACEUTICS REVIEW(S)

**OFFICE OF CLINICAL PHARMACOLOGY REVIEW**

| | |
|---|---|
| BLA: | 761046 |
| Submission Date(s): | 11/23/2015 |
| Relevant IND: | 012823 |
| Brand Name: | To Be Determined |
| Generic Name: | Bezlotoxumab |
| Primary Reviewer: | Yang He, Ph.D. |
| Team Leader: | Seong H. Jang, Ph.D. |
| PM Reviewer: | Luning Zhuang, Ph.D. |
| PM Team Leader: | Jeffry Florian, Ph.D. |
| OCP Division: | DCP4 |
| OND Division: | DAIP |
| Applicant: | Merck Sharp & Dohme Corp., a subsidiary of Merck & Co., Inc. (Merck) |
| Submission Type: | Original Biologics License Application (BLA) |
| Formulation; Strength(s): | Injection solution for intravenous use; 1000 mg/40 mL (25 mg/mL) in a single vial |
| Indication | Prevention of *Clostridium difficile* infection (CDI) recurrence in patients 18 years or older receiving antibiotic therapy for CDI |

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## 1. EXECUTIVE SUMMARY

The current theory of pathogenesis of *Clostridium difficile* infection (CDI) is that toxigenic strains of *C. difficile*, either endogenous to the colon or recently acquired, flourish after disruption of the normal bacterial colonic flora and release toxins A and B that target the gut epithelium leading to pathogenic changes. In vitro studies have shown that antibodies against toxin A and toxin B can prevent binding of the toxins to their target cells and neutralize cytotoxic activities of these toxins. Bezlotoxumab was developed as a fully human monoclonal antibody (mAb) of the IgG1/kappa isotype subclass that binds with high affinity to *C. difficile* toxin B. It prevents binding of toxin B to its target cells, blocking the cellular intoxication cascade at its first step, and averting the damage and inflammation that lead to the symptoms of CDI. Actoxumab is a mAb targeted against *C. difficile* toxin A, which was hypothesized to play a role in prevention of CDI recurrence when combined with bezlotoxumab. An IND was initiated for a combination of actoxumab and bezlotoxumab (MK-3415A, IND 012823) on 12/30/2005, and Fast Track designation was granted to MK-3415A for the treatment of patients with CDI and/or for the prevention of CDI recurrence. However, based on overall assessment of efficacy and safety data, the Sponsor seeks approval for bezlotoxumab intravenous injection alone. The proposed indication is for the prevention of *Clostridium difficile* infection (CDI) recurrence in patients 18 years or older receiving antibiotic therapy for CDI. The recommended dose of bezlotoxumab is 10 mg/kg administered as an intravenous (IV) infusion over 60 minutes as a (b) (4) dose.

The pharmacokinetics (PK), safety, and immunogenicity of bezlotoxumab were characterized in four Phase 1 trials (Studies P020, P004, P005, and P006). The safety and efficacy of bezlotoxumab administered concomitantly with antibiotics for CDI treatment were evaluated in two Phase 2 trials (Studies P017 and P018) and two pivotal Phase 3 trials (Studies P001 and P002), in which a total of 1664 patients with a diagnosis of CDI (CDI patients) was included. The dose of bezlotoxumab evaluated in these studies was 10 mg/kg administered intravenously, and no dose ranging study was conducted to evaluate bezlotoxumab efficacy in CDI patients. Population PK and exposure-response analyses were conducted using pooled data from three Phase 1 trials (Studies P004, P005, and P006) and two Phase 3 trials (Studies P001 and P002), since the bioanalytical methods used in the initial Phase 1 and 2 studies (Studies P020, P017, and P018) was not specific enough to measure bezlotoxumab concentration in serum.

Geometric mean clearance (CL) of bezlotoxumab is 0.317 L/day, geometric mean volume of distribution (Vd) is 7.33 L, and elimination half-life (t½) is approximately 19 days in CDI patients, which is similar to other human monoclonal antibodies typically having a low CL and a limited Vd. Bezlotoxumab shows moderate PK variability. The %CV for geometric mean of $AUC_{0\text{-inf}}$ and $C_{max}$ was 40% and 21%, respectively. The estimated inter-individual variability in CL and central volume of distribution (Vc) was approximately 28.7% and 10.6%, respectively. Bezlotoxumab is not expected to specifically bind to serum proteins. As a monoclonal antibody, bezlotoxumab is degraded into small peptides and individual amino acids through protein catabolism. Bezlotoxumab can be detected in stools from a limited number of CDI patients after IV administration. No dose adjustment is needed for intrinsic and extrinsic factors when the weight-based dose is administered.

The overall incidence of AEs and SAEs in the bezlotoxumab and actoxumab + bezlotoxumab groups was comparable to the placebo group. There was no clinically meaningful exposure-response relationship for efficacy or safety identified to support a need of dose adjustment to address efficacy and safety concerns. Single dose of bezlotoxumab did not result in the development of anti-bezlotoxumab antibodies in serum. No comparability issue for drug product as well as drug substance was identified.

### 1.1. Recommendation

The Clinical Pharmacology results submitted in this BLA appear sufficient and appropriate to support approval of bezlotoxumab intravenous injection for the proposed indication of prevention of CDI recurrence in patients 18 years or older receiving antibiotic therapy for CDI. From a Clinical Pharmacology perspective, we recommend approval of this BLA.

### 1.2. Phase 4 Commitments

No phase IV commitments are recommended.

### 1.3. Summary of Important Clinical Pharmacology Findings

The clinical pharmacology characteristics of bezlotoxumab have been studied in healthy subjects and CDI patients. These studies show bezlotoxumab demonstrates the following clinical pharmacology characteristics:

- General PK characteristics
  Following single intravenous administration of bezlotoxumab 10 mg/kg in CDI patients, the mean clearance (CL) of bezlotoxumab is 0.317 L/day, with a volume of distribution (Vd) of 7.33 L, and an elimination half-life (t½) of approximately 19 days. The clearance value is much smaller than the glomerular filtration rate ($\geq$ 90 mL/min/1.73m$^2$) in subjects with normal renal function, indicating minimum contribution of renal excretion to the total bezlotoxumab clearance. The volume of distribution value is within the range of volumes of serum and extracellular water, indicating limited extravascular distribution and/or tissue binding. These findings are consistent with PK characteristics of other human monoclonal antibodies, which typically have low clearance and small volume of distribution. Bezlotoxumab has moderate PK variability (40% and 21% CV for $AUC_{0\text{-inf}}$ and $C_{max}$, respectively). In the Population PK (PopPK) analysis, the inter-subject variability in CL and Vc was estimated at approximately 28.7% and 10.7%, respectively.

- Distribution, metabolism and excretion
  Bezlotoxumab is not expected to specifically bind to serum proteins. As a monoclonal antibody, bezlotoxumab is degraded into small peptides and individual amino acids through protein catabolism. Thus, bezlotoxumab is not expected to be metabolized by the liver or excreted by the kidney, although no mass balance study was conducted in humans. Bezlotoxumab is not a substrate of hepatic metabolic enzymes/transporters. The

target of bezlotoxumab is an exogenous toxin but not a cytokine modulator. Therefore, bezlotoxumab is not expected to be an inhibitor or an inducer of metabolic enzymes/transporters. Bezlotoxumab was detected in stools in ≤16% of CDI patients who gave stool samples after the bezlotoxumab administration.

- Effect of intrinsic/extrinsic factors
  The PopPK analysis of serum bezlotoxumab concentrations from the Phase 1 and Phase 3 trials demonstrated no clinical relevant effects of renal impairment and hepatic impairment on bezlotoxumab PK. In addition, the PopPK analysis showed that other intrinsic or extrinsic factors (i.e. albumin, gender, age, race/ethnicity, clinical comorbidities, CDI severity, and concomitant use of non-standard care of antibiotics) would not affect the exposure of bezlotoxumab to a clinically meaningful extent. Hence, beyond the weight-based dose, no dose adjustments are required for intrinsic or extrinsic factors.

- Exposure-response (E-R) relationships for efficacy and safety
  There was no apparent relationship between serum exposure to bezlotoxumab and clinical efficacy endpoints. The E-R analyses for safety, including adverse events (AEs) and serious adverse events (SAEs), indicated that patient covariates (i.e. albumin, concomitant use of non-standard of care systemic antibiotics or PPIs, hospitalization, Charlson Comorbidity Index ≥ 3, and age), rather than exposure, are the primary factors influencing the incidence of AEs and SAEs. In addition, there were no substantial differences in the incidence of each category of AEs/SAEs between the placebo and the bezlotoxumab treatment arms. Overall, there was no E-R relationship for efficacy or safety identified to support a need for dose adjustment to address efficacy and safety concerns (b) (4)
  (b) (4)
  (b) (4)

- Immunogenicity
  In healthy subjects and CDI patients, no bezlotoxumab treatment-emergent anti-drug antibody against bezlotoxumab has been observed following a single 10 mg/kg bezlotoxumab administration, indicating that there may be very limited immunogenicity due to bezlotoxumab.

## 2. QUESTION BASED REVIEW

It has been shown that actoxumab and bezlotoxumab neutralize the cytotoxic activities of toxins (Toxins A and B, respectively) from a broad range of clinical isolates of *C. difficile* in nonclinical studies. However, the clinical development program has demonstrated that bezlotoxumab significantly reduces recurrence of CDI compared to placebo, while actoxumab + bezlotoxumab do not have any safety or efficacy benefit over bezlotoxumab alone. Actoxumab alone is not efficacious. The Sponsor selected bezlotoxumab as the final product for registration. Thus, this Clinical Pharmacology review focuses on clinical data related to bezlotoxumab.

### 2.1. General Attributes of the Drug

*2.1.1. What are the highlights of the chemistry and physical-chemical properties of the drug substance and the formulation of the drug product?*

**Drug Substance:**

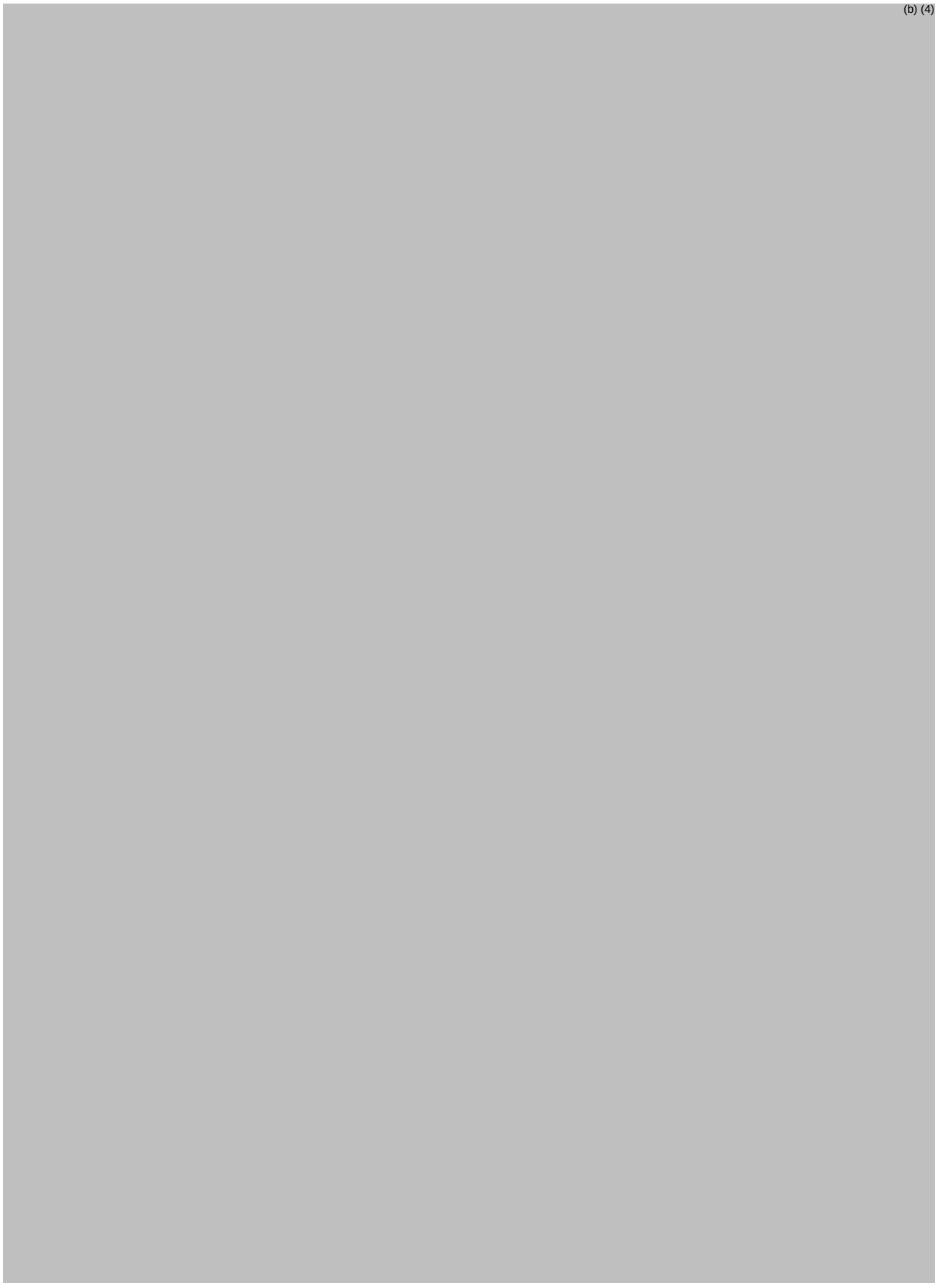
Bezlotoxumab belongs to the IgG1/kappa isotype subclass. **Table 2.1.1-1** shows the nomenclature that has been used for bezlotoxumab drug substance over the course of product development. The theoretical molecular weights of the (b) (4) (b) (4).

**Table 2.1.1-1** Nomenclature for Bezlotoxumab Drug Substance

| | |
|---|---|
| **Code Name** | MK-6072 |
| **Other Code Name** | MDX-1388, (b) (4) |
| **CAS Registry Number** | 1246264-45-8 |
| **CAS Name** | anti-(*Clostridium difficile* Toxin B) Immunoglobulin G1 (human monoclonal γ-chain), disulfide with human monoclonal κ-chain, dimer |
| **Generic Name: rINN** | bezlotoxumab |
| **Generic Name: USAN** | bezlotoxumab |
| **Trade Name** | To be determined |

CAS: Chemical Abstracts Service
rINN: Recommended International Nonproprietary Name
USAN: United States Adopted Name

Protein Structure:
Bezlotoxumab is an IgG1 monoclonal antibody (b) (4) (b) (4)


(b) (4)

**Drug Product**

Bezlotoxumab drug product (DP) is a sterile, aqueous, preservative-free solution (b) (4) filled into vials for single use. **Table 2.1.1-2** shows composition of bezlotoxumab DP. Each vial contains a target deliverable dose of 40 mL bezlotoxumab at 25 mg/mL for a total of 1000 mg per vial. The product is supplied in a 50-mL (b) (4) (b) (4) vial (b) (4) (b) (4) For administration, bezlotoxumab DP is diluted with either 0.9% (w/v) sodium chloride or 5% (w/v) dextrose, and the resulting admixture infusion solution is administered intravenously.

**Table 2.1.1-2** Composition of Bezlotoxumab Drug Product

| Ingredient | Compendial Grade | Target Amount (mg/vial) [a] | Target Concentration | Function |
|---|---|---|---|---|
| **MK-6072** | NA | 1000 | 25 mg/mL | Active ingredient |
| **Sodium Chloride** | *Ph. Eur./USP* | (b) (4) | | (b) (4) |
| **Sodium Citrate Dihydrate**[b] | *Ph. Eur./USP* | | | |
| **Citric Acid Monohydrate**[b] | *Ph. Eur./USP* | | | |
| **Polysorbate 80** | *Ph. Eur./NF* | | | |
| **DTPA** | *USP* [c] | | | |
| **Water for Injection** | *Ph. Eur./USP* | (b) (4) | | |

[a] (b) (4)

[b]

[c] DTPA is listed in the *USP* as pentetic acid. There is no *Ph.Eur.* monograph for this material.

DTPA: diethylenetriaminepentaacetic acid
NA: not applicable
qs: "quantum sufficit", the quantity that is sufficient

Bezlotoxumab clinical DP has been manufactured by (b) (4) (b) (4) and by Merck Sharp and Dohme (Merck; West Point, Pennsylvania, U.S.). Commercial supply manufacture is performed at MSD Ireland (Carlow). *In vitro* analytical evaluation was conducted to assess the process changes implemented during development and comparability of all materials manufactured for use in nonclinical and clinical studies. The Sponsor concluded that materials manufactured by the different processes were comparable.

*2.1.2. What is the proposed mechanism of drug action and therapeutic indication?*

Bezlotoxumab is a fully human IgG1 monoclonal antibody that neutralizes Toxin B by binding to the combined repetitive oligopeptide (CROP) domains at regions that partially overlap with the putative receptor (carbohydrate) binding pockets, thereby preventing toxin binding to the cell

surface. Blocking of this step in the intoxication cascade prevents the downstream damage and inflammation that underlie *Clostridium difficile* infection (CDI). Thus, bezlotoxumab does not have antimicrobial activity and does not take the place of antibiotic therapy for CDI. The Sponsor proposed that bezlotoxumab is expected to prevent recurrent infections after the antibiotic treatment is stopped, by providing passive immunity against *C. difficile* toxin B produced by the outgrowth of persistent or newly acquired spores, thereby preventing new or further damage to the gut epithelium.

The preclinical data indicated that bezlotoxumab crossed the gut wall through paracellular transport, enhanced by toxin-induced disruption of the gut epithelium, to reach the site of infection in the lumen of the gut. In addition, toxin may leak into the sub-epithelial space of the gut wall through the same mechanism. Thus, neutralization of the toxin by bezlotoxumab may occur both on the luminal and systemic sides of the intestinal wall.

The proposed indication is for prevention of CDI recurrence in patients 18 years or older receiving antibiotic therapy for CDI.

*2.1.3. What is the proposed dosage and route of administration?*

The recommended dose of bezlotoxumab is 10 mg/kg administered as an intravenous (IV) infusion over 60 minutes as a single dose. The required volume is withdrawn from the vial(s) based on the patient's weight (kg) and transferred into an IV bag containing 0.9% Sodium Chloride Injection or 5% Dextrose Injection to prepare a diluted solution with a final concentration ranging from 1 to 10 mg/mL.

## 2.2. General Clinical Pharmacology

*2.2.1. What are the design features of the clinical pharmacology and clinical studies used to support dosing or claims?*

The clinical pharmacology program included four Phase 1 trials evaluating the safety, PK, and immunogenicity of bezlotoxumab, administered alone or in combination (actoxumab + bezlotoxumab), in healthy adult subjects. These trials containing bezlotoxumab included (**Table 2.2.1-1**): a dose-ranging PK trial of bezlotoxumab and actoxumab alone and in combination (Study P020), a trial assessing a 60 minute infusion of actoxumab + bezlotoxumab to support a shorter infusion duration in the Phase 3 program (Study P005), a trial of actoxumab + bezlotoxumab in Japanese subjects (Study P006), and a trial examining the immunogenicity and tolerability of a second dose of actoxumab+bezlotoxumab given 12 weeks after the first dose (Study P004). These assessments evaluated initial safety/tolerability and PK over a broad range of doses from 0.3 mg/kg to 20 mg/kg of bezlotoxumab alone or in combination with actoxumab, administered as a single IV infusion over 60-120 minutes. Across these four Phase 1 trials of bezlotoxumab alone or in combination with actoxumab, 138 subjects, including 52 females were enrolled. Thirty subjects received bezlotoxumab alone, 96 received both bezlotoxumab and actoxumab, and 12 received placebo.

The comprehensive bezlotoxumab Phase 2/3 clinical development program included four trials in which bezlotoxumab and actoxumab were studied alone or in combination (actoxumab + bezlotoxumab). As summarized in **Table 2.2.1-1**, the two Phase 2 trials were Studies P018 and P017, and the two Phase 3 trials were Studies P001 and P002. Each of these clinical trials was a randomized, double-blind, placebo-controlled, multi-center, safety, and efficacy trial conducted in adults 18 years of age or older who were concurrently receiving standard of care (SoC) antibiotic treatment for a primary or recurrent episode of CDI. The trials were designed to assess whether a single IV infusion of the mAbs (either alone or in combination), given with standard of care antibiotics, decreases the proportion of subjects with CDI recurrence, as compared to treatment with a single infusion of placebo given with standard of care antibiotics. The two pivotal Phase 3 trials followed subjects for efficacy and safety for 12 weeks; they were identical in design, conduct, and statistical analysis with the following exceptions:

(1) Study P002 had 3 treatment groups instead of 4 (the actoxumab alone arm was not included in the trial because in the earlier Phase 2 trial (Study P018), the CDI recurrence rate among subjects who had received actoxumab alone was similar to the rate observed in the subjects who had received placebo);

(2) Study P002 did not include a planned interim analysis for stopping enrollment in the individual mAb treatment groups; and

(3) Study P002 had an extended follow-up period through Month 12 in a subset of subjects (approximately 300) to assess for CDI recurrence and colonization with toxigenic *C. difficile*.

**Table 2.2.1-1** Summary of Clinical Trials of Actoxumab and Bezlotoxumab

| Trial | Phase | Protocol Title |
|---|---|---|
| CA-CDA1-04-01 (Protocol 019) | 1 | Open-Label, Dose Escalation Phase 1 Study in Healthy Volunteers to Evaluate the Safety and Pharmacokinetics of *Clostridium difficile* Toxin A Human Monoclonal Antibody (CDA1[†]) |
| CA-GCDX-05-01 (Protocol 020) | 1 | Open-Label, Dose Escalation Phase 1 Study in Healthy Volunteers to Evaluate the Safety and Pharmacokinetics of *Clostridium difficile* Toxin A Human Monoclonal Antibody (GS CDA1[†]) and *Clostridium difficile* Toxin B Human Monoclonal Antibody (MDX-1388[‡]) |
| Protocol 005 | 1 | A Single Dose Study to Evaluate the Safety, Tolerability, and Pharmacokinetics of a 1 hour Intravenous Infusion of MK 3415A[§] |
| Protocol 006 | 1 | A Single Dose Study to Evaluate the Safety, Tolerability, and Pharmacokinetics of MK-3415A[§] in Healthy Japanese Male Subjects |
| Protocol 004 | 1 | A Study to Evaluate the Immunogenicity of MK-3415A[§] |
| CA-CDA1-05-02 (Protocol 018) | 2 | A Phase 2 Randomized, Double-Blind, Placebo-Controlled Study of the Clinical Effectiveness of a Human Monoclonal Antibody to Toxin A (CDA1[†]) in Patients being Treated for *Clostridium difficile* Associated Diarrhea (CDAD) |
| CA-GCDX-06-02 (Protocol 017) | 2 | A Phase 2 Randomized, Double-Blind, Placebo-Controlled Study of the Clinical Effectiveness of a Human Monoclonal Antibody to *Clostridium difficile* Toxin A (GS-CDA1[†]) and a Human Monoclonal Antibody to *Clostridium difficile* Toxin B (MDX-1388[‡]) in Patients being Treated for *Clostridium difficile* Associated Disease |
| Protocol 001 | 3 | A Phase 3, Randomized, Double-Blind, Placebo Controlled, Adaptive Design Study of the Efficacy, Safety, and Tolerability of a Single Infusion of MK-3415[†], MK-6072[‡], and MK-3415A[§] in Patients Receiving Antibiotic therapy for CDI. |
| Protocol 002 | 3 | A Phase 3, Randomized, Double-Blind, Placebo Controlled Study of the Efficacy, Safety, and Tolerability of a Single Infusion of MK-6072[‡] and MK-3415A[§] in Patients Receiving Antibiotic therapy for CDI. |

[†]Actoxumab, which is also referred to as CDA1, GS-CDA1, or MK-3415.
[‡]Bezlotoxumab, which is also referred to as MDX-1388 or MK-6072.
[§]The combination of actoxumab + bezlotoxumab, which is also referred to as MK-3415A.

The Phase 3 clinicFigure al development program includes 781 subjects who received bezlotoxumab alone, 773 subjects who received the combination of actoxumab + bezlotoxumab, and 773 subjects who received placebo in the efficacy analysis population (the Full Analysis Set, FAS, population) across the two pivotal clinical trials. Similarly, 786 subjects who received bezlotoxumab alone, 777 subjects who received the combination of actoxumab + bezlotoxumab, and 781 subjects who received placebo were included in the safety population across the 2 trials.

In addition, PopPK and exposure-response analyses were conducted with pooled data from the Phase 1 (Studies P004, P005, and P006) and Phase 3 trials (Studies P001 and P002) that used an ECL assay specific for bezlotoxumab to quantify bezlotoxumab serum concentrations (see *Section 2.2.3*).

*2.2.2. What is the basis for selecting the response endpoints (i.e., clinical or surrogate endpoints) or biomarkers (collectively called pharmacodynamics [PD]) and how are they measured in clinical pharmacology and clinical studies?*

The primary efficacy endpoint in the pivotal Phase 3 trials was the proportion of subjects in the full analysis set (FAS) population with CDI recurrence during the 12 week (Day 85 ± 5 days) follow-up period. CDI recurrence was defined as the development of a new episode of diarrhea associated with a positive stool test either from the local or central laboratory for toxigenic *C. difficile* following clinical cure of the baseline episode. CDI recurrence typically occurs within 8 to 10 weeks following an initial CDI episode. Clinical cure was defined as no diarrhea for 2 consecutive days following a ≤14 day regimen of SoC antibiotic. Therefore, the follow-up period of 12 weeks for the Phase 3 studies was specifically chosen to ensure that CDI recurrences would not be missed. This efficacy endpoint reflects the proposed indication.

In the two pivotal Phase 3 studies, enrolled patients 18 years of age or older and who had a confirmed diagnosis of CDI, which was defined as diarrhea (passage of 3 or more loose bowel movements in 24 or fewer hours) and a positive stool test for toxigenic *C. difficile* from a stool sample collected no more than 7 days before study entry. Patients were excluded if surgery for CDI was planned, or if they had uncontrolled chronic diarrheal illness. Patients received a 10- to 14-day course of oral SoC antibiotic for CDI (metronidazole, vancomycin or fidaxomicin) and a single infusion of bezlotoxumab or placebo was administered prior to completion of the SoC antibiotic therapy, and the patients were followed for 12-weeks after the infusion. The Sponsor proposed efficacy assessment was based on the primary efficacy endpoint in the FAS population (**Table 2.2.2-1**). See Clinical and Statistical Reviews for assessment of the efficacy analysis results.

**Table 2.2.2-1: CDI Recurrence Rate through 12 Weeks after Infusion (Full Analysis Set [*])**

| Bezlotoxumab with SoC [†] Percent (n/N) | Placebo with SoC [†] Percent (n/N) | Adjusted Difference (95% CI) [‡] | p-value |
|---|---|---|---|
| 16.5 (129/781) | 26.6 (206/773) | -10.0 (-14.0, -6.0) | <0.0001 |

n = Number of subjects in the analysis population meeting the criteria for endpoint

N = Number of subjects included in the analysis population

[*] Full Analysis Set = a subset of all randomized subjects with exclusions for: (i) did not receive infusion of study medication, (ii) did not have a positive local stool test for toxigenic *C. difficile*; (iii) did not receive protocol defined standard of care therapy within a 1 day window of the infusion

[†] SoC = Standard of Care antibiotic (metronidazole or vancomycin or fidaxomicin)

[‡] One sided p-value based on the Miettinen and Nurminen method stratified by protocol (Studies P001 and P002), SoC antibiotic (metronidazole vs. vancomycin vs. fidaxomicin) and hospitalization status (inpatient vs. outpatient)

*2.2.3. Are the active moieties in the biological fluid appropriately identified and measured to assess pharmacokinetic parameters?*

Development of bioanalytical methods for measuring bezlotoxumab concentration in human serum included two stages.

In an initial Phase 1 trial (Study P020) and a Phase 2 trial (Study P017), bezlotoxumab concentration measurements and anti-drug antibody (ADA) assessments in serum were performed using a ligand-capture-based enzyme-linked immunosorbent assay (ELISA). This assay cannot distinguish endogenous anti-toxin B antibodies and bezlotoxumab.

Subsequent Phase 1 clinical trials (Studies P004, P005, and P006) and the pivotal Phase 3 trials (Studies P001 and P002) employed a conventional sandwich electrochemiluminescence (ECL) immunoassay for the quantitation of the serum concentration of bezlotoxumab, which was specific for bezlotoxumab. The active moiety bezlotoxumab was appropriately identified and measured in serum from humans.

***2.2.4. Exposure-Response***

*2.2.4.1. What are the characteristics for exposure-response relationships (dose-response, concentration-response) for efficacy?*

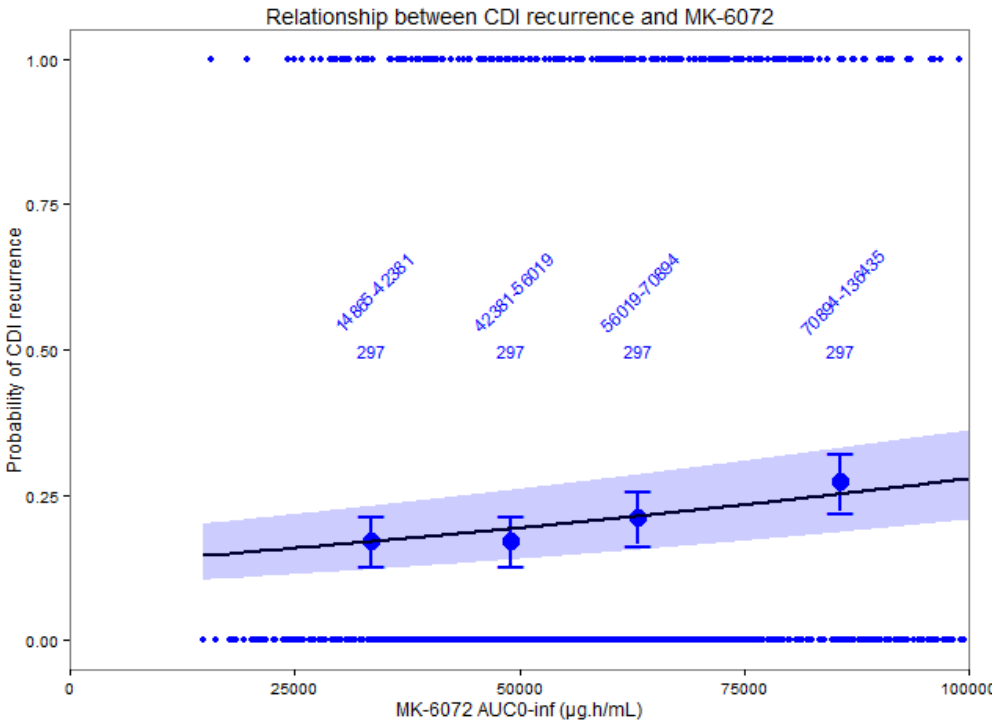
The exposure-response relationship for efficacy of bezlotoxumab has been evaluated in CDI patients in the Phase 3 studies. Both Studies P001 and P002 were randomized, multicenter, double-blind, placebo-controlled, trials conducted in subjects 18 years of age or older who were receiving standard of care treatment for a primary or recurrent episode of CDI. The primary

efficacy endpoint in these pivotal Phase 3 trials was the proportion of subjects in the FAS population with CDI recurrence during the 12-week (Day 85 ± 5 days) follow-up period after a clinical cure of the baseline CDI episode. The CDI recurrence was defined as development of a new episode of diarrhea (3 or more loose stools in 24 hours) associated with a positive stool test for toxigenic *C. difficile.*

The Sponsor's E-R analysis evaluated potential relationships between serum exposure of bezlotoxumab and clinical efficacy endpoints. The Reviewers have concerns about the selected exposure and efficacy variables in the Sponsor's E-R analysis. Ideally, the efficacy of bezlotoxumab may be more related to the concentration in intestine than serum concentration because the major action (binding to Toxin B) site of bezlotoxumab is likely to be in the intestine and/or gut epithelium and the lumen of intestine. In addition, the distribution of bezlotoxumab into the aforementioned sites of action in CDI patients may be affected by the integrity of the gut wall and, thus, is not likely to be correlated directly with serum exposure. Furthermore, the Reviewers found that the Sponsor's proposed E-R relationship primarily focused on the pre-determined efficacy endpoint of CDI recurrence. This endpoint embedded a component mechanistically unrelated to drug effect (i.e. positive culture of toxigenic *C. difficile* in stool). Thus, the Sponsor proposed E-R relationship between bezlotoxumab serum exposure and CDI recurrence was not considered to be valid to evaluate the proposed bezlotoxumab dose of 10 mg/kg single IV infusion. However, because the measurement of bezlotoxumab in stool was reported qualitatively in limited CDI patients from a Phase 3 study (Study P002), the Reviewer evaluated potential relationships between bezlotoxumab serum exposure or patient's covariates and the proposed efficacy endpoint for completeness.

In the Reviewer's pharmacometrics analysis, a relationship was identified between serum exposure and CDI recurrence with higher exposure associated with a higher rate of CDI recurrence in patients who experienced clinical initial cure during SoC (**Figure 2.2.4.1-1**, see Section 3.2 Pharmacometrics Review for details). This unusual observation indirectly supports that the efficacy of bezlotoxumab is not related with serum exposure (see above). The logistic regression analysis showed that several patient covariates had a significant impact on the probability of CDI recurrence, including baseline albumin (note: albumin lab values were only available at baseline and week 4), history of CDI in the past 6 months, age, and a Charlson Comorbidity Index ≥ 3. Since albumin is highly correlated with bezlotoxumab exposure (AUC) and also associated with the patient health, the relationship between CDI recurrence and bezlotoxumab serum exposure is confounded with baseline albumin.

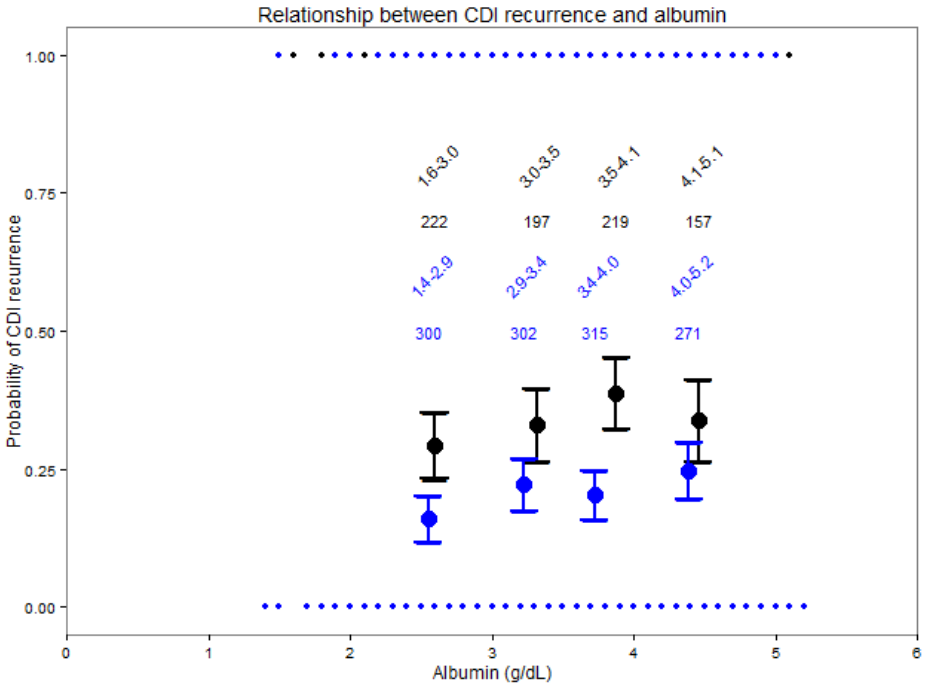
**Figure 2.2.4.1-1: Relationship between CDI recurrence and bezlotoxumab (MK-6072)**




*The number with ranges on the top represent the bezlotoxumab exposure ranges for each quartile and the numbers under the ranges represent the number of patients of each quartile.*
*Note: The reviewer used data from bezlotoxumab containing treatment and placebo arms in the Phase 3 trials (n=1188), excluding the patients who experienced clinical initial failure during standard of care. Source: Reviewer's Pharmacometrics analysis (**Section 3.2**)*

To better understand the relationship between albumin and CDI recurrence, the Reviewer plotted the CDI recurrence versus four quartiles of baseline albumin as shown in **Figure 2.2.4.1-2** for both treatment and placebo. Overall, CDI recurrence increases with higher albumin concentrations in both treatment arms. One advantage of looking at albumin in this manner is that it permits a direct quartile-to-quartile comparison of the odds ratio between the treatment and placebo arms. While an overall trend in response with respect to albumin may be observed within a treatment arm, if the same trend is also observed in the placebo arm this suggests it is patient characteristics that are contributing factors to the relationship and not bezlotoxumab exposure. As shown in **Table 2.2.4.1-1**, the odds ratio between placebo and treatment are relatively consistent across all quartiles, supporting a consistent treatment effect across all quartiles. The results suggest that the exposure provided by 10 mg/kg bezlotoxumab approach a flat response rate regarding CDI recurrence and that higher exposure is not predicted to produce further benefit treatment benefit. Therefore, no dose adjustment is necessary based on this observation.

**Figure 2.2.4.1-2: Relationship between CDI recurrence and albumin**




*Blue depicts the treatment arm and black depicts the placebo arm. The numbers with ranges on the top represent the albumin concentration ranges for each quartile and the numbers under the ranges represent the number of patients of each quartile.*
*Note: The reviewer used dataset from bezlotoxumab containing treatment and placebo arms in the Phase 3 trials (n=1983). Source: Reviewer's Pharmacometrics analysis (**Section 3.2**)*

**Table 2.2.4.1-1: Odds ratio of CDI recurrence for the treatment arm compared to the placebo arm across the four quantiles of albumin**

| Albumin quantile | 1st quantile | 2nd quantile | 3rd quantile | 4th quantile |
|---|---|---|---|---|
| Odds ratio | 0.55 | 0.67 | 0.52 | 0.73 |

*Source: Reviewer's Pharmacometrics analysis*

*2.2.4.2. What are the characteristics for exposure-response relationships (dose-response, concentration-response) for safety?*

There were no apparent differences in the overall adverse event rate, severe adverse event rate, or types of adverse events observed between the placebo and the bezlotoxumab-containing treatment arms in the Phase 3 studies (i.e., Studies P001 and P002). One or more adverse events (AEs) were reported in 61.7% and 61.2% patients of the bezlotoxumab and placebo arms, respectively.

Serious adverse events were slightly more frequent in the placebo arm (32.7%) compared to the bezlotoxumab arm (29.4%). The most frequent serious adverse events were gastrointestinal disorders and infections and infestations (**Table 2.2.4.2-1**). The Reviewer found that adverse

events could include diarrhea, which as discussed above, was one of the components of the efficacy analysis. As such, the imbalance in serious adverse events favoring bezlotoxumab may reflect bezlotoxumab's effect on reducing the occurrence of diarrhea associated with CDI.

**Table 2.2.4.2-1** Subjects with Serious Adverse Events (>3%) during 12 Weeks Following Infusion in the Phase 3 Studies P001 and P002

| | (Pooled) Actoxumab/ Bezlotoxumab | (Pooled) Bezlotoxumab | (Pooled) Placebo |
|---|---|---|---|
| Total subjects No. | 777 | 786 | 781 |
| With one or more serious adverse events | 212 (27.3%) | 231 (29.4%) | 255 (32.7%) |
| Cardiac disorders | 24 (3.1%) | 36 (4.6%) | 27 (3.5%) |
| Gastrointestinal disorders | 37 (4.8%) | 49 (6.2%) | 42 (5.4%) |
| Infections and infestations | 93 (12.0%) | 104 (13.2%) | 138 (17.7%) |
| Nervous system disorders | 24 (3.1%) | 13 (1.7%) | 8 (1.0%) |
| Respiratory, thoracic and mediastinal disorders | 26 (3.3%) | 28 (3.6%) | 24 (3.1%) |

Source: Applicant's summary of clinical safety report, Page 377, Appendix 2.7.4: 16 (Adapted)

There were no substantial differences in the incidence of each category of AEs/SAEs between the placebo and the bezlotoxumab treatment arms and, thus, there were no specific AEs/SAEs warranted exploration of the relationship with bezlotoxumab serum exposure. Accordingly, additional exposure-response analyses for specific AEs/SAEs were not conducted.

Considering the systemic effect of bezlotoxumab and the pathology of CDI, the evaluation of exposure-response relationship between bezlotoxumab and overall AEs/SAEs, as the Sponsor conducted, is not considered to be meaningful because (1) some of AEs (e.g., administration site conditions) may not be relevant to systemic exposure of bezlotoxumab and (2) including diarrhea as a measurement of AEs may mislead the exposure-response analysis of safety (see above). However, the Reviewers conducted an additional sensitivity analysis to evaluate the difference between treatment arm and placebo arm by quantile of baseline albumin concentrations, the result is in line with the observation of similar SAEs between bezlotoxumab treatment and placebo arms (**Section 3.2** Pharmacometrics Review). Overall, no exposure-adverse event relationship was identified to support a need of dose adjustment to address any safety concern.

*2.2.4.3. What is the effect of bezlotoxumab on QTc interval?*

A mAb has a low likelihood of direct ion channel interactions. It is typically not associated with clinically meaningful effects on QTc interval owing to large size, which prevents interaction with the pore of hERG channels. In addition, there is no evidence from nonclinical or clinical data to suggest that bezlotoxumab has the potential to delay ventricular repolarization. Thus, a dedicated QTc trial was not performed for bezlotoxumab. In the later Phase 1 trials (Studies P004, P005, and P006), however, ECG monitoring and time-matched PK sampling were assessed after administration of 10 mg/kg or 20 mg/kg of actoxumab + bezlotoxumab. In a PK/QTc analysis,

there was a trend of decreasing change from baseline population-corrected QT interval with increasing bezlotoxumab serum concentration that was statistically significant but is not clinically meaningful. Similarly, no clinically significant effect on QTc interval was observed after administration of bezlotoxumab alone or actoxumab + bezlotoxumab (Studies P001 and P002). Furthermore, a graphical analysis of bezlotoxumab serum concentrations and QTc interval revealed no trends for QTc prolongation with increasing bezlotoxumab concentration. Hence, across healthy subjects and patients, bezlotoxumab does not affect QTc interval as reflected by change from baseline.

*2.2.4.4. Is the dose and dosing regimen selected by the applicant consistent with the known relationship between dose-concentration-response, and are there any unresolved dosing or administration issues?*

Only one dose level was evaluated in Phase 3 studies by the Sponsor. No dose ranging trials for efficacy were conducted beyond Phase 1 studies. The exposure-response analyses using the Phase 3 data show that no relationship was identified between exposure and efficacy/safety based on one dose level. A higher dose (20 mg/kg) was also administered to a limited number of healthy subjects in several Phase 1 trials. Although no dose-related toxicities were observed after the 20 mg/kg dose in healthy subjects, efficacy and safety of a lower or higher dose in CDI patients are unknown.

*2.2.5. What are the PK characteristics of bezlotoxumab?*

The pharmacokinetics of bezlotoxumab following intravenous administration were evaluated in healthy subjects and CDI patients**.** Most of the clinical trials including the trials in the Clinical Pharmacology program were conducted with actoxumab + bezlotoxumab as described in *Section 2.2.1*. There are no analytic incompatibilities (see *Section 2.6*) and no PK interaction between actoxumab and bezlotoxumab (see *Sections 2.2.5.1* and *2.4.2.6*); hence, the PK of bezlotoxumab can be considered independently of actoxumab in subjects who received both mAbs.

*2.2.5.1. What are the single dose and multiple dose PK parameters?*

***Single Dose Pharmacokinetics of Bezlotoxumab***

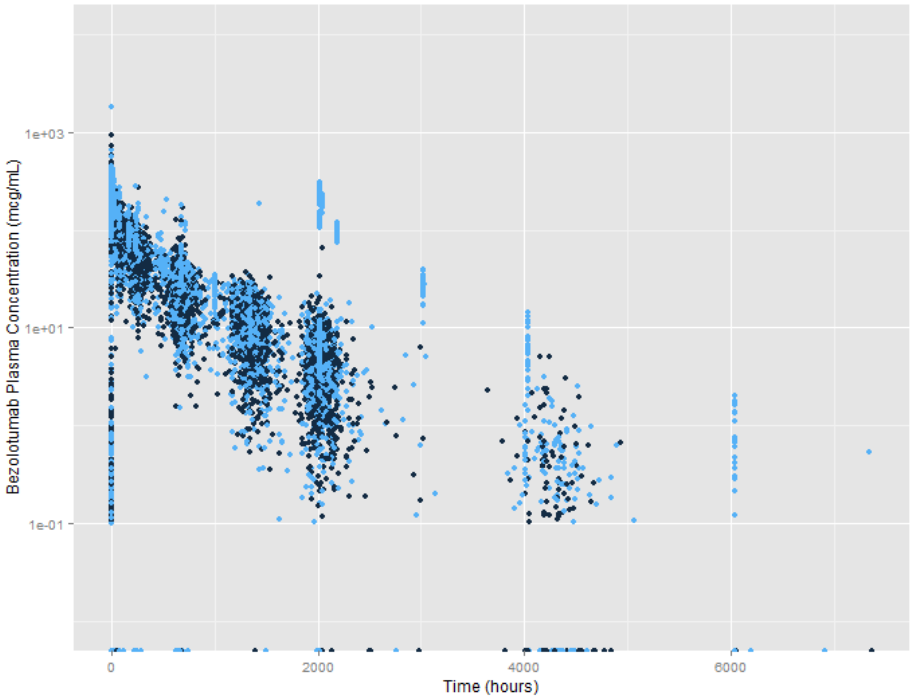
The Phase 1 study P020 is the only clinical study evaluating bezlotoxumab PK following administration of single doses ranging from 0.3 mg/kg to 20 mg/kg in healthy adult subjects (see *Section 2.2.5.8*). As stated by the Sponsor, however, the bioanalytical assay used for measuring bezlotoxumab in this study is not specific to distinguish endogenous anti-toxin B antibodies and bezlotoxumab. Thus, the PK data obtained from this study (Study P020) is excluded from the final bezlotoxumab PK assessment.

Characterization of bezlotoxumab PK is supported by a PopPK analysis based on the data obtained from three Phase 1 (Studies P004, P005, and P006) and two Phase 3 (Studies P001 and P002) trials. These trials used the ECL assay for the evaluation of actoxumab and bezlotoxumab

concentrations. Intensive PK sampling was obtained in healthy subjects in the Phase 1 trials. A dense PK sampling design of at least six PK samples per subject was performed in the Phase 3 trials.

The PopPK analysis dataset includes 72 healthy subjects in the Phase 1 trials who received bezlotoxumab alone or actoxumab + bezlotoxumab (Studies P004, P005, and P006), including 29 subjects who received a second dose of actoxumab + bezlotoxumab (Study P004). Following a single dose of 10 mg/kg, the PK characteristics of bezlotoxumab in healthy subjects are similar across all three Phase 1 studies. Please refer to *Section 2.2.5.2* for more detail of bezlotoxumab PK in healthy subjects. The PK characteristics of bezlotoxumab in patients were evaluated in a PopPK analysis, and this PopPK dataset includes 1515 patients in the Phase 3 program (Studies P001 and P002) who received a 10 mg/kg dose of bezlotoxumab alone or 10 mg/kg actoxumab + 10 mg/kg bezlotoxumab (henceforth noted as 10 mg/kg actoxumab + bezlotoxumab). The scatter plot of concentration-time profiles shows similar PK profiles of bezlotoxumab following administration of 10 mg/kg dose of bezlotoxumab or actoxumab + bezlotoxumab in patients **(Figure 2.2.5.1-1)**. In addition, no evidence of target-mediated drug disposition was observed based on visual inspection of individual concentration time profiles (i.e., no accelerated elimination at lower concentrations).

**Figure 2.2.5.1-1**. Concentration-Time Profiles for Bezlotoxumab in Patients Following Administration of 10 mg/kg Bezlotoxumab (black dots) or 10 mg/kg Actoxumab + Bezlotoxumab (blue dots)




Based on the PopPK analysis, the geometric mean (%CV) clearance (CL) of bezlotoxumab is 0.317 L/day (40%), with a volume of distribution (Vd) of 7.33 L (16%), and an elimination half-

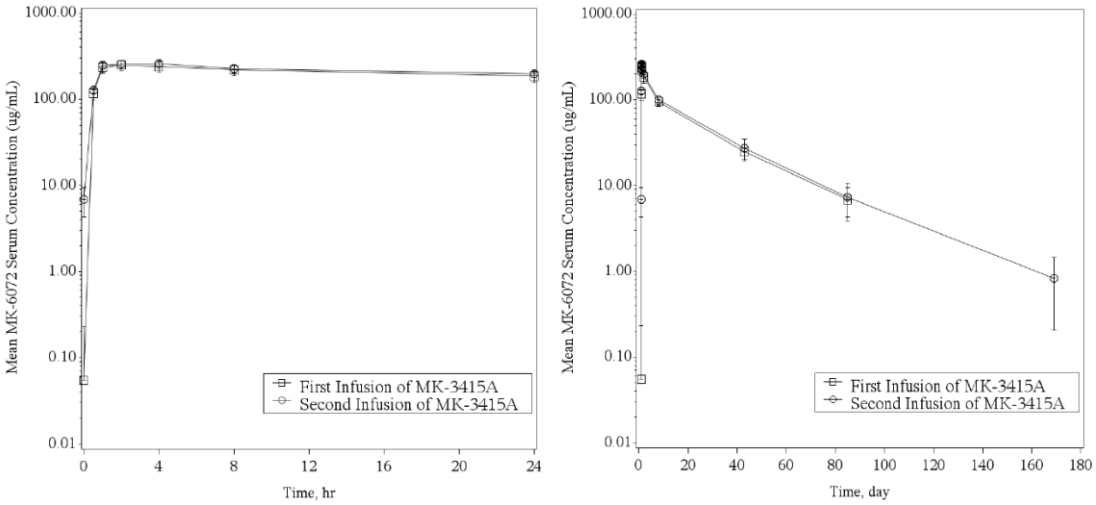
life ($t_{½}$) of approximately 19 days (28%) in patients. In patients who received a single 10 mg/kg IV dose of bezlotoxumab, mean bezlotoxumab $AUC_{0\text{-}inf}$ and $C_{max}$ are 53,000 µg/mL and 185 µg/mL, respectively. Bezlotoxumab shows moderate PK variability (40% and 21% CV for $AUC_{0\text{-}inf}$ and $C_{max}$, respectively).

***Multiple Dose Pharmacokinetics of Bezlotoxumab***

Although bezlotoxumab is intended for single dose administration, the Sponsor conducted Study P004 to evaluate the immunogenicity, safety and PK of actoxumab and bezlotoxumab after two doses of 10 mg/kg of each actoxumab and bezlotoxumab administered 84 days (12 weeks) apart.

The mean serum concentration-time profiles of bezlotoxumab following administration of MK-3415A (a combination of actoxumab and bezlotoxumab) as a single 1-hour infusion to healthy adult subjects are presented below in **Figure 2.2.5.1-2**. **Table 2.2.5.1-1** presents the statistical summary of bezlotoxumab serum pharmacokinetics in healthy adult subjects following administration of MK-3415A (10 mg/kg of each mAb, actoxumab and bezlotoxumab) as 2 consecutive single 1-hour infusions, separated by 84 days. Overall, bezlotoxumab exhibited similar pharmacokinetics following the second infusion relative to the first infusion of 10 mg/kg of MK-3415A. Minimal accumulation in bezlotoxumab exposure was observed when the second dose is administered 84 days after the first dose.

**Figure 2.2.5.1-2**. Mean (± SD) Serum Concentration-Time Profiles of bezlotoxumab Following the First and Second Infusion of MK-3415A in Healthy Adult Subjects (N = 30 First Infusion/N = 29 Second Infusion) (Left, First 24 Hours Postdose; Right, Entire Sampling Interval)




MK-6072: Bezlotoxumab (Source: Study Report P004)

**Table 2.2.5.1-1**. Statistical Comparison of Serum Pharmacokinetics of bezlotoxumab Following the Administration of 10 mg/kg MK-3415A via a 1-Hour IV Infusion in Healthy Adult Subjects

| | | First Infusion of MK-3415A | | | Second Infusion of MK-3415A | | | Second Infusion/ First Infusion | | Pseudo Within Subject %CV[‖] |
|---|---|---|---|---|---|---|---|---|---|---|
| Monoclonal Antibody | Pharmacokinetic Parameter | N | GM | 95% CI | N[††] | GM | 95% CI | GMR | 90% CI | |
| MK-6072 | AUC0-∞[†] (µg/mL•hr) | 30 | 85700 | (80500, 91200) | 29 | 91300 | (84700, 98400) | 1.07 | (1.04, 1.10) | 6.249 |
| | AUC0-84 days[†] (µg/mL•hr) | 30 | 81200 | (76700, 85900) | 29 | 85400 | (80100, 91200) | 1.05 | (1.03, 1.08) | 5.772 |
| | Cmax[†] (µg/mL) | 30 | 250 | (238, 264) | 29 | 261 | (251, 271) | 1.04 | (1.02, 1.07) | 5.409 |
| | Vss[†] (mL) | 30 | 5381.65 | (5052.29, 5732.48) | 29 | 5312.16 | (4940.45, 5711.83) | | | |
| | CL[†] (mL/hr) | 30 | 8.57 | (7.89, 9.31) | 29 | 8.05 | (7.36, 8.81) | | | |
| | Tmax[‡] (hr) | 30 | 2.00 | (1.00, 4.08) | 29 | 2.01 | (1.00, 4.02) | | | |
| | Apparent terminal $t_{½}$[¶] (hr) | 30 | 474.46 | 15.42 | 29 | 541.80 | 21.27 | | | |

First Infusion (Day 1): Single IV dose of MK-3415A (10 mg/kg of each mAb, MK-3415 and MK-6072) as a 1-hour infusion.
Second Infusion (Day 85): Single IV dose of MK-3415A (10 mg/kg of each mAb, MK-3415 and MK-6072) as a 1-hour infusion.
[†]Back-transformed least-squares mean and confidence interval from linear mixed-effects model performed on natural log-transformed values.
[‖]Pseudo within-subject %CV = 100*Sqrt[($\sigma^2_A$ + $\sigma^2_B$ - 2*$\sigma_{AB}$)/2], where $\sigma^2_A$ and $\sigma^2_B$ are the estimated variances on the log scale for the 2 treatment infusions, and $\sigma_{AB}$ is the corresponding estimated covariance, each obtained from the linear mixed-effects model.
[‡]Median (min, max) reported for Tmax.
[¶]Geometric mean and percent CV reported for apparent terminal $t_{½}$.
[††]Subject AN 0023 was lost to follow-up and did not receive the second infusion. Subject AN 0023's AUC0-84 days value was excluded from statistical analysis for first infusion, as subject had no blood draw on Day 84 (his exposure represents data through Day 43 only) following first infusion.
GM = Geometric least-square mean; GMR = Geometric least-square mean ratio; CI = Confidence interval
AUC0-84 days was added as an additional endpoint as the dosing interval was 84 days.

*2.2.5.2. How does the PK of the drug and its major active metabolites in healthy volunteers compare to that in patients?*

Based on the PopPK analysis result using the PK data from three Phase 1 trials (Studies P004, P005, and P006) and two Phase 3 trials (Studies P001 and P002), bezlotoxumab $C_{max}$ and $AUC_{0\text{-}inf}$ in healthy subjects following a 10 mg/kg dose of bezlotoxumab or actoxumab + bezlotoxumab (**Table 2.2.5.2-1)** are 24.3% and 43% higher than those in CDI patients following a 10 mg/kg dose (**Table 2.2.5.2-2**), respectively. The Sponsor provided an explanation that the observed exposure difference between healthy subjects and patients is due to 43% higher serum albumin concentrations in healthy subjects than CDI patients, and a strong relationship between albumin concentration and CL and Vc was observed in the PopPK analysis. It has been reported that albumin and IgG antibodies are both protected from lysosomal degradation by the same Neonatal Fc receptor (FcRn) recycling system, though they interact with different binding sites. Changes in the capacity of FcRn protection can influence concentrations of albumin and IgG antibodies in a similar fashion, thus serum levels of albumin and IgG antibodies increase and decrease concomitantly. Consistently, bezlotoxumab exposure was found to positively correlate with baseline albumin concentration in the PopPK analysis. Please refer to the Pharmacometrics Review for more details (**Section 3.2**). It should be noted that the PK parameters (i.e. AUC0-inf, AUC0-84d, and Cmax) were calculated after actual doses of 8-12 mg/kg in the clinical trials and not adjusted to align with the standard dose of 10 mg/kg. Thus, these reported PK parameters may contain wider ranges of variability, compared to those after the standard dose of 10 mg/kg.

**Table 2.2.5.2-1.** Summary of Bezlotoxumab PK Parameter Values in **Healthy Subjects** (n = 65) Following Administration of a Single IV Dose of 10 (± 20%) mg/kg of Bezlotoxumab or 10 (± 20%) mg/kg Actoxumab + Bezlotoxumab, based on Population PK Analysis with the data obtained from three Phase 1 trials (Studies P004, P005, and P006).

| | AUC0-inf (µg.h/mL) | AUC0-84d (µg.h/mL) | Cmax (µg/mL) | t1/2 (day) | Tmax† (h) | CL (L/day) | Vd (L) |
|---|---|---|---|---|---|---|---|
| Geometric mean | 75800 | 70300 | 230 | 22.9 | . | 0.237 | 6.91 |
| Geometric CV% | 17.8 | 15.1 | 14.0 | 15.1 | . | 21.4 | 10.0 |
| Median | 75700 | 70300 | 231 | 22.5 | 1.00 | 0.237 | 6.85 |
| 10th percentile | 62300 | 59700 | 190 | 19.1 | 0.97 | 0.183 | 6.22 |
| 90th percentile | 95800 | 86700 | 280 | 27.6 | 1.06 | 0.312 | 7.79 |

Based on posthoc exposures for healthy subjects who received actual doses between 8 – 12 mg/kg
†Only Median and Range reported

**Table 2.2.5.2-2**. Summary of Bezlotoxumab PK Parameter Values in the **Patient Population** (n = 1504) Following Administration of a Single IV Dose of 10 (± 20%) mg/kg of Bezlotoxumab or 10 (± 20%) mg/kg Actoxumab + Bezlotoxumab, based on Population PK Analysis with the data obtained from two Phase 3 trials (Studies P001 and P002)

| | AUC0-inf (µg.h/mL) | AUC0-84d (µg.h/mL) | $C_{max}$ (µg/mL) | t1/2 (day) | Tmax† (h) | CL (L/day) | Vd (L) |
|---|---|---|---|---|---|---|---|
| Geometric mean | 53000 | 50400 | 185 | 18.7 | . | 0.317 | 7.33 |
| Geometric CV% | 40.2 | 36.7 | 20.7 | 28.0 | . | 40.6 | 16.3 |
| Median | 54700 | 52300 | 184 | 18.9 | 1.00 | 0.309 | 7.36 |
| 10th percentile | 31700 | 31500 | 144 | 13.1 | 0.50 | 0.195 | 5.96 |
| 90th percentile | 85600 | 77800 | 244 | 26.2 | 4.73 | 0.533 | 8.98 |

Based on posthoc exposures for patients who received actual doses between 8 – 12 mg/kg
†Only Median and Range reported

### *2.2.5.3. What are the characteristics of drug absorption?*

Description of absorption characteristics is not applicable, as bezlotoxumab is formulated for intravenous administration.

### *2.2.5.4. What are the characteristics of drug distribution?*

Consistent with a limited extravascular distribution and/or tissue binding of typical human mAbs, the mean of volume of distribution of bezlotoxumab is 7.33 L (16% CV) based on the PopPK analysis, and is within the range of the volumes of serum and extracellular water. Bezlotoxumab is not expected to specifically bind to serum proteins.

### *2.2.5.5. Does the mass balance study suggest renal or hepatic as the major route of elimination?*

A mass balance study was not conducted for bezlotoxumab.

### *2.2.5.6. What are the characteristics of drug metabolism?*

As a monoclonal antibody, bezlotoxumab is expected to be degraded into small peptides and individual amino acids through protein catabolism.

*2.2.5.7. What are the characteristics of drug excretion?*

In general, monoclonal antibodies are not excreted via renal elimination. The renal excretion of bezlotoxumab is consistent with other human mAbs. Based on qualitative analysis results (positive/negative) in CDI patients from the Phase 3 study (Study P002), bezlotoxumab was detected in stools in ≤16% of CDI patients who gave stool samples after the bezlotoxumab administration (**Table 2.2.5.7-1**). It should be noted that some patients had positive bezlotoxumab in stools even before the drug administration, so this result should be interpreted with caution.

**Table 2.2.5.7-1** Percentage of Stool Samples with Detectable Bezlotoxumab (MK-6072) at Each Visit by Treatment (Study P002)

| Treatment | Nominal Day | Detectable | Non-Detectable | QNS[a] |
|---|---|---|---|---|
| MK-3415A | 1-Pre-infusion | 0/48 ( 0.0) | 47/48 (97.9) | 1/48 (2.1) |
| | 1-Post-infusion | 47/222 (21.2) | 171/222 (77.0) | 4/222 (1.8) |
| | 4 | 54/162 (33.3) | 108/162 (66.7) | 0/162 (0.0) |
| | 11 | 24/159 (15.1) | 134/159 (84.3) | 1/159 (0.6) |
| | 29 | 28/141 (19.9) | 112/141 (79.4) | 1/141 (0.7) |
| | Unscheduled[b] | 34/133 (25.6) | 98/133 (73.7) | 1/133 (0.8) |
| MK-6072 | 1-Pre-infusion | 3/50 ( 6.0) | 44/50 (88.0) | 3/50 (6.0) |
| | 1-Post-infusion | 16/211 ( 7.6) | 191/211 (90.5) | 4/211 (1.9) |
| | 4 | 21/134 (15.7) | 111/134 (82.8) | 2/134 (1.5) |
| | 11 | 11/137 ( 8.0) | 126/137 (92.0) | 0/137 (0.0) |
| | 29 | 8/115 ( 7.0) | 107/115 (93.0) | 0/115 (0.0) |
| | Unscheduled[b] | 11/127 ( 8.7) | 116/127 (91.3) | 0/127 (0.0) |

a. Quantity not sufficient, could not be analyzed.
b. The listed sample numbers in the unscheduled row may reflect more than one sample per subject.

*2.2.5.8. Based on PK parameters, what is the degree of linearity or nonlinearity in the dose-concentration relationship?*

A Phase 1 single ascending dose study (Study P020) is the only clinical study assessing dose-concentration relationship with a series of dose levels; however, the bioanalytical assay used in this study was not able to specifically detect bezlotoxumab in serum. Instead, it measured a combination of endogenous anti-toxin B IgG and bezlotoxumab. Thus, the study result is not valid to evaluate linearity of bezlotoxumab PK. For purpose of reference, the $AUC_{0\text{-inf}}$ of bezlotoxumab in healthy subjects across the 0.3 to 20 mg/kg dose range from this study is summarized in **Table 2.2.5.8-1**.

**Table 2.2.5.8-1** Summary of Bezlotoxumab Pharmacokinetic Parameters in Healthy Subjects (N=6 in Each Cohort)
(NOTE: Nonspecific bioanalytical assay for bezlotoxumab quantification was used. This table is provided for reference ONLY.)

| MDX-1388 Dose | 0.3 mg/kg | 1 mg/kg | | 3 mg/kg | | 10 mg/kg | | 20 mg/kg | |
|---|---|---|---|---|---|---|---|---|---|
| | Alone | Alone | + 1 mg/kg GS-CDA1 | Alone | + 10 mg/kg GS-CDA1 | Alone | + 10 mg/kg GS-CDA1 | Alone | + 20 mg/kg GS-CDA1 |
| **Cmax (µg/mL)** | | | | | | | | | |
| Mean | 6.91 | 33.31 | 24.46 | 67.59 | 85.57 | 223.31 | 270.69 | 513.20 | 500.21 |
| SD | 0.93 | 11.91 | 2.93 | 10.74 | 19.44 | 19.81 | 52.67 | 71.07 | 78.24 |
| Minimum | 5.85 | 22.91 | 21.47 | 50.91 | 64.30 | 201.16 | 203.31 | 450.34 | 393.84 |
| Median | 6.64 | 28.16 | 23.68 | 70.22 | 84.99 | 225.86 | 260.75 | 500.03 | 482.08 |
| Maximum | 8.22 | 50.26 | 28.23 | 80.87 | 105.99 | 253.58 | 344.88 | 623.39 | 620.55 |
| **Tmax (hr)** | | | | | | | | | |
| Mean | 3.67 | 1.09 | 1.08 | 2.25 | 16.01 | 1.67 | 1.34 | 6.56 | 2.93 |
| SD | 2.94 | 0.78 | 0.49 | 2.91 | 17.91 | 1.83 | 2.31 | 8.19 | 2.75 |
| Minimum | 0.02 | 0.07 | 0.50 | 0.00 | 0.50 | 0.00 | 0.00 | 0.50 | 0.50 |
| Median | 3.00 | 1.00 | 1.00 | 0.50 | 13.00 | 0.76 | 0.50 | 4.00 | 2.28 |
| Maximum | 8.00 | 2.00 | 2.00 | 6.00 | 44.55 | 4.00 | 6.00 | 22.83 | 6.00 |
| **t½z (hr)** | | | | | | | | | |
| Mean | 612.94 | 595.75 | 651.28 | 493.40 | 523.83 | 664.46 | 512.06 | 529.99 | 458.39 |
| SD | 265.36 | 123.49 | 378.04 | 110.25 | 107.25 | 223.81 | 92.77 | 203.16 | 74.42 |
| Minimum | 473.09 | 414.94 | 294.89 | 398.15 | 406.83 | 336.55 | 361.47 | 237.07 | 337.10 |
| Median | 512.97 | 599.80 | 566.40 | 462.32 | 503.69 | 655.81 | 539.62 | 494.38 | 473.03 |
| Maximum | 1152.65 | 746.54 | 1369.52 | 700.41 | 684.97 | 1014.92 | 618.31 | 849.01 | 547.77 |
| **$AUC_{(0\text{-}tlast)}$** | | | | | | | | | |
| Mean | 2229.53 | 8538.41 | 8236.37 | 21567.99 | 23773.54 | 73578.69 | 78315.88 | 173781.02 | 144885.80 |
| SD | 263.88 | 1530.51 | 2351.59 | 2558.01 | 3629.44 | 11064.46 | 11411.60 | 68035.77 | 17650.94 |
| Minimum | 1844.81 | 6215.95 | 5728.24 | 18842.24 | 16460.20 | 61811.71 | 63232.76 | 92365.85 | 121685.02 |
| Median | 2201.01 | 8642.93 | 7881.51 | 21483.58 | 24858.86 | 74299.82 | 81803.44 | 164165.71 | 142999.42 |
| Maximum | 2665.36 | 10608.96 | 11724.40 | 25157.30 | 26236.49 | 91886.50 | 89283.47 | 298038.72 | 172439.18 |
| **$AUC_{(0\text{-}\infty)}$** | | | | | | | | | |
| Mean | 2626.74 | 9359.43 | 9896.87 | 23147.80 | 26082.58 | 84568.80 | 84406.52 | 192991.78 | 153784.69 |
| SD | 668.08 | 1723.54 | 4200.35 | 3289.50 | 4689.80 | 13443.29 | 14105.58 | 62097.57 | 20832.34 |
| Minimum | 2073.47 | 6395.53 | 5855.87 | 19568.32 | 16912.73 | 63120.96 | 66333.12 | 118249.41 | 124627.81 |
| Median | 2418.69 | 9536.52 | 8558.50 | 22451.49 | 27160.64 | 84754.95 | 89649.70 | 185644.26 | 154864.47 |
| Maximum | 3955.80 | 11343.81 | 15567.51 | 27803.02 | 30379.31 | 101730.24 | 97157.35 | 305639.71 | 182932.64 |

MDX-1388: bezlotoxumab; GS-CDA1: actoxumab (Source: Study P020)

### *2.2.5.9. How do the PK parameters change with time following chronic dosing?*

Bezlotoxumab is intended for single dose administration. A Phase 1 study (Study P004) was conducted with two IV infusions on Day 1 and Day 85. The PK of bezlotoxumab following the two IV infusions was very similar.

### *2.2.5.10. What is the inter- and intra-subject variability of PK parameters in volunteers and patients, and what are the major causes of variability?*

Bezlotoxumab has moderate PK variability (40% and 21% CV for $AUC_{0\text{-}inf}$ and $C_{max}$, respectively). In the PopPK analysis, the inter-subject variability in CL and Vc was estimated as approximately 28.7% and 10.7%, respectively. Residual variability was estimated as 62%.

The effects of body weight/BMI, albumin, gender, age, race/ethnicity, renal impairment, hepatic impairment, clinical comorbidities, and CDI severity on the exposure of bezlotoxumab were assessed with a PopPK analysis of serum bezlotoxumab concentrations from the Phase 1 and Phase 3 trials. Evaluation of subject covariates demonstrated that body weight, gender, and albumin were statistically significant covariates contributing to variability in bezlotoxumab PK (see *Section 2.3*).

*2.2.5.11. Does the clinical pharmacology information presented by the applicant support the use of a 10 mg/kg bezlotoxumab dose for prevention of CDI?*

The recommended dose for bezlotoxumab is a single 10 mg/kg dose administered via an IV infusion over 60 minutes. During the drug development process, a single infusion of actoxumab + bezlotoxumab at 10 mg/kg each was evaluated in a Phase 2 trial (P017), and this dose showed a significant reduction of CDI recurrence and was generally well tolerated. The Sponsor indicated that higher doses were thought unlikely to demonstrate a clinically meaningful benefit and lower doses were not studied in Phase 3 trials, as acceptable safety was demonstrated at 10 mg/kg in the Phase 1 and Phase 2 trials. Thus, no dose-ranging study has been conducted to evaluate efficacy or safety for bezlotoxumab. In the pivotal Phase 3 trials, a reduction in the rate of CDI recurrence was observed after the administration of 10 mg/kg bezlotoxumab, compared to placebo. No safety findings were observed that would suggest a lower dose would have an improved risk-benefit profile.

*2.2.5.12. What clinical pharmacology information was used to evaluate clinical significance of the changes in the exposure of bezlotoxumab due to intrinsic or extrinsic factor?*

Because there are several concerns in the E-R analysis (see *Section 2.2.4*), the results of the E-R analysis for efficacy and safety cannot be used to evaluate clinical significance of the changes in the exposure of bezlotoxumab due to intrinsic or extrinsic factors. The Sponsor introduced a "comparability bounds" to evaluate clinical significance of the changes in the exposure of bezlotoxumab due to intrinsic or extrinsic factors. It refers to a range of bezlotoxumab pharmacokinetic exposures, relative to those achieved at a clinical dose of a single 10 mg/kg IV infusion of bezlotoxumab in patients within the Phase 3 trials (Studies P001 and P002), which has been demonstrated to have clinical comparability with respect to the safety and efficacy of bezlotoxumab. When observed or predicted changes in the pharmacokinetic exposure of bezlotoxumab fall within these pre-specified comparability bounds, safety and efficacy are expected to be comparable to that of the recommended clinical dose of a single 10 mg/kg IV infusion of bezlotoxumab. $AUC_{0\text{-}inf}$ was selected as the exposure indicator to define the comparability bound. The comparability bounds of (0.6, 1.6) for bezlotoxumab are determined as the ratios of $10^{th}$ (31,700 μg·h/mL) and $90^{th}$ (85,600 μg·h/mL) percentiles to the median (54,700 μg·h/mL) of observed bezlotoxumab $AUC_{0\text{-}inf}$ values, respectively, following administration of a single 10 mg/kg IV infusion of bezlotoxumab alone or as actoxumab + bezlotoxumab in the Phase 3 trials. These $10^{th}$ and $90^{th}$ percentiles of observed $AUC_{0\text{-}inf}$ are consistent with the PopPK modeling result. The $10^{th}$ percentile of $AUC_{inf}$ following the single 10 mg/kg dose showed similar efficacy (i.e. CDI recurrence rate) to the median $AUC_{inf}$. This dose of single 10 mg/kg had acceptable safety and tolerability in the Phase 3 trials. Accordingly, the Reviewers agree that this comparability bounds of (0.6, 1.6) for changes in bezlotoxumab $AUC_{0\text{-}inf}$ is appropriate and can be used as a criterion to evaluate the need of potential dose adjustment in a given patient subpopulation.

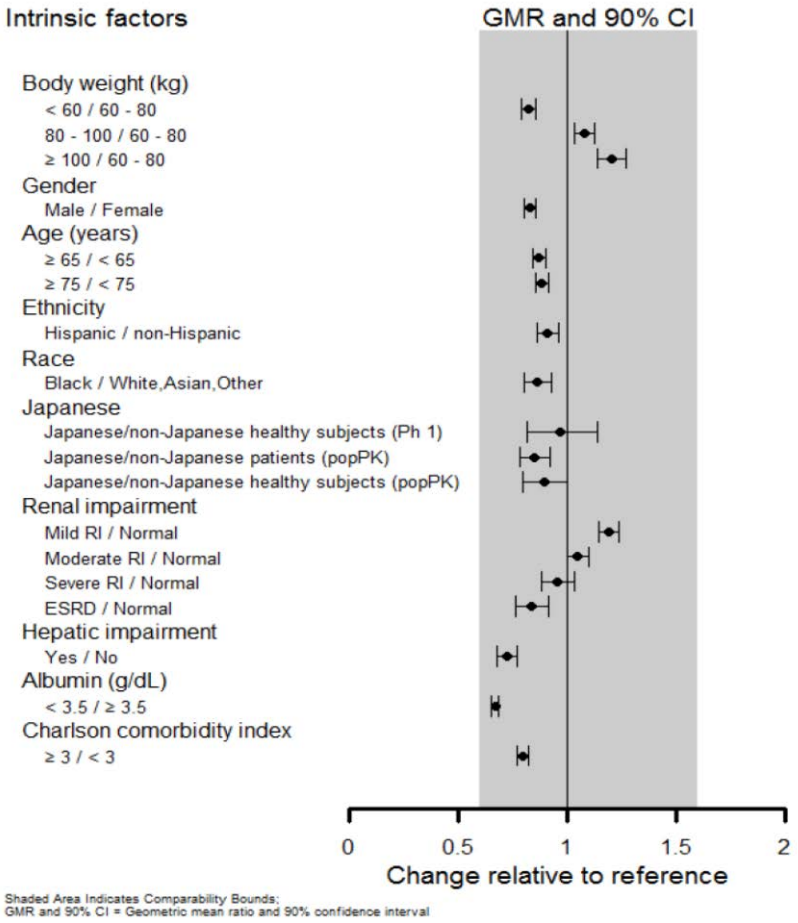
## 2.3. Intrinsic Factors

*2.3.1. What intrinsic factors influence exposure and/or response, and what is the impact of any differences in exposure on efficacy or safety responses?*

In general, therapeutic antibodies are primarily eliminated by protein catabolism, and clearance is not dependent on a single organ. Consequently, intrinsic factors such as organ dysfunction or age typically have limited effect on the exposure of therapeutic antibodies and, therefore, are not anticipated to affect the exposure of bezlotoxumab to a clinically meaningful extent. Dedicated Phase 1 trials of intrinsic factors and organ dysfunction were not conducted for bezlotoxumab, except for a single trial (Study P006) investigating the PK of bezlotoxumab in healthy Japanese subjects.

The effects of body weight/BMI, albumin, gender, age, race/ethnicity, renal impairment, hepatic impairment, clinical comorbidities, and CDI severity on the exposure of bezlotoxumab were assessed with a PopPK analysis of serum concentrations from the Phase 1 and Phase 3 trials. Body weight was included as a covariate on clearance and volume of distribution terms as part of the structural model, and covariate effects for baseline albumin, gender, race, and Japanese origin on clearance or volume of distribution were included in the final PopPK model. Overall, the PopPK analysis confirmed the expectation that intrinsic or extrinsic factors did not affect the exposure of bezlotoxumab to a clinically meaningful extent, with changes in bezlotoxumab $AUC_{0\text{-}inf}$ estimates within the comparability bounds (0.6, 1.6) relative to the reference groups. Hence, beyond the weight-based dose, no dose adjustments are required. A forest plot summarizing the key exposure results by intrinsic factor derived from the PopPK analysis is provided in **Figure 2.3.1-1**, with the shaded region denoting the comparability bounds of (0.6, 1.6).

**Figure 2.3.1-1** Effect of Intrinsic Factors on Bezlotoxumab $AUC_{0\text{-inf}}$ Based on PopPK Analysis

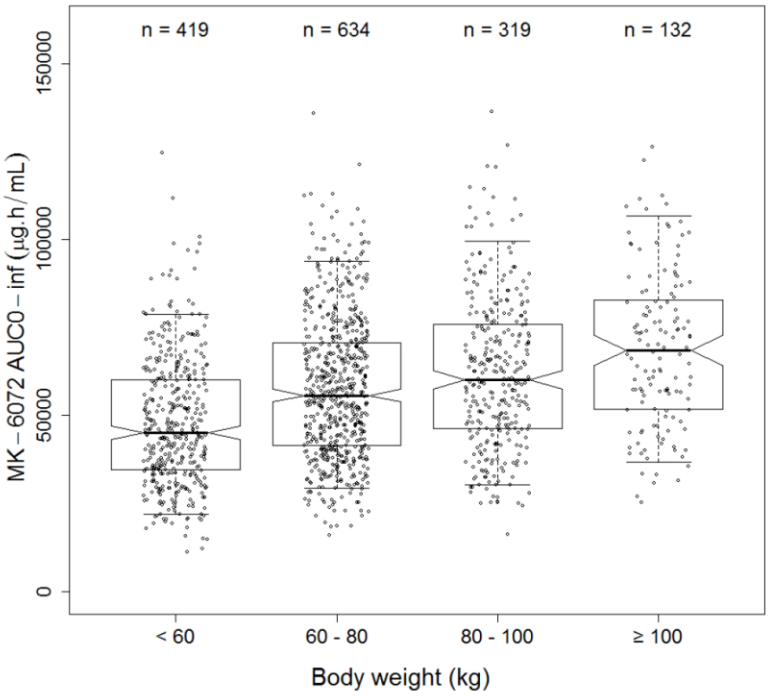



*2.3.2. Based upon what is known about exposure-response relationships and their variability and the groups studied, healthy volunteers vs. patients vs. specific populations (examples shown below), what dosage regimen adjustments, if any, are recommended for each of these groups? If dosage regimen adjustments are not based upon exposure-response relationships, describe the alternative basis for the recommendation.*

*2.3.2.1. Weight*

As anticipated based on experience with other monoclonal antibodies, weight is a significant covariate for bezlotoxumab PK, specifically on CL and Vc. The incremental impact of body weight across the intended target patient population is limited with a 18% decrease (in $AUC_{0\text{-inf}}$ for patients weighing <60 kg and a 20% increase in $AUC_{0\text{-inf}}$ for patients weighing ≥ 100 kg compared to patients in the 60 – 80 kg range, as depicted in **Figure 2.3.2.1-1**.

**Figure 2.3.2.1-1** Distribution of Bezlotoxumab $AUC_{0\text{-}inf}$ in Patients Following Administration of a Single IV Dose of 10 mg/kg Bezlotoxumab or 10 mg/kg Actoxumab + Bezlotoxumab Across Body Weight Categories



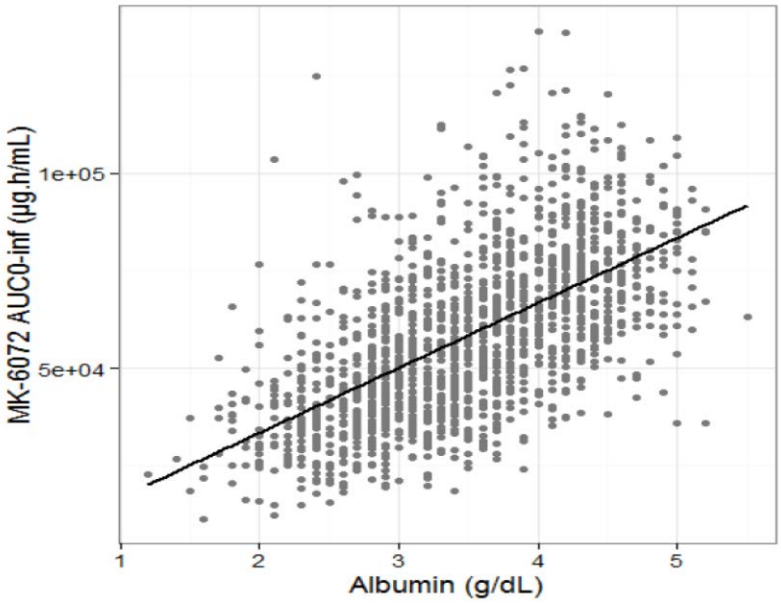

MK-6072 = bezlotoxumab
Note: The boxes represent 25th and 75th percentiles; the line reflects the median; the whiskers are 5th and 95th percentiles.

Overall, assessment of the effect of body weight demonstrated that the comparative fold change in AUC consistently falls within the comparability bounds (0.6, 1.6), indicating that a dose adjustment beyond that implemented in the clinical trials is not needed. Therefore, the recommended dose of 10 mg/kg can be given regardless of body weight.

*2.3.2.2. Albumin*

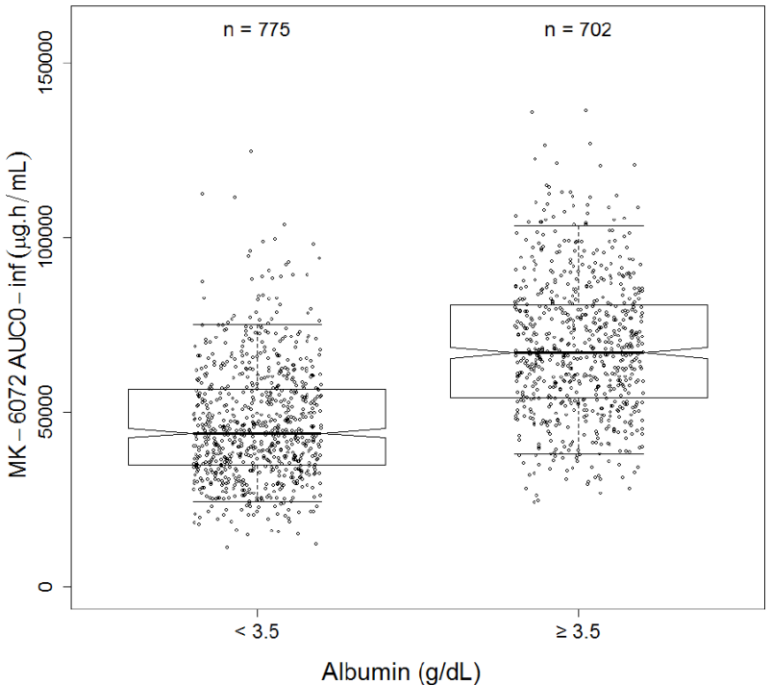
It is suggested that albumin and antibodies are protected from lysosomal degradation by the same FcRn recycling system, though they interact with different binding sites. Factors that affect the capacity of FcRn recycling influence concentrations of albumin and IgG antibodies in a similar fashion, so that serum levels of albumin and IgG antibodies increase and decrease concomitantly. Formal testing of the covariate relationship indicates that clearance and central volume of distribution decrease with increasing albumin concentrations. **Figure 2.3.2.2-1** shows the relationship of bezlotoxumab exposure and albumin concentrations in serum.

**Figure 2.3.2.2-1** Association of Bezlotoxumab $AUC_{0\text{-inf}}$ with Baseline Albumin Concentrations in Patients and Healthy Subjects (N = 1542) Following Administration of a Single IV Dose of 10 mg/kg Bezlotoxumab or 10 mg/kg Actoxumab + Bezlotoxumab each




Bezlotoxumab exposures were 33% lower in patients with albumin concentrations < 3.5 g/dL compared to patients with normal albumin concentrations (**Figure 2.3.2.2-2**). In patients with below-normal albumin concentrations, the changes in exposure fall within the clinical experience that defines the comparability bounds of (0.6, 1.6), as indicated in **Figure 2.3.1-1**. Albumin concentrations correlate with covariates related to patient health such as the Charlson Comorbidity Index, Zar score, and hospitalization, consistent with patients in poorer health having lower albumin concentrations. However, the Reviewer's subgroup analyses using data from the Phase 3 trials showed that CDI recurrence rate was comparable regardless of albumin concentration. Thus, the effect of albumin on bezlotoxumab $AUC_{0\text{-inf}}$ is not clinically meaningful and no dose adjustment is warranted.

**Figure 2.3.2.2-2** Distribution of Bezlotoxumab $AUC_{0\text{-}inf}$ in Patients with Abnormal (< 3.5 g/dL) and Normal (≥ 3.5 g/dL) Albumin Concentration Following Administration of a Single IV Dose of 10 mg/kg Bezlotoxumab or 10 mg/kg Actoxumab + Bezlotoxumab each (MK-3415A)




*2.3.2.3. Elderly*

The age range for subjects included in the population analysis was 18 to 100 years. No influence of age on CL or Vc of bezlotoxumab was identified from the population pharmacokinetic analysis. Thus, no dose adjustments based on age are recommended.

*2.3.2.4. Pediatric patients*

Bezlotoxumab has not been studied in children (individuals under 18 years of age). As agreed in the amended initial Pediatric Study Plan submitted to IND 12,823 on 11/06/2015, pediatric assessments were deferred until after approval of the BLA in adults. Accordingly, the Sponsor has not proposed dosing recommendations for pediatric patients.

*2.3.2.5. Gender*

In the PopPK analysis, gender is a significant covariate on clearance and central volume of distribution, consistent with other mAbs. Bezlotoxumab $AUC_{0\text{-}inf}$ was 18% lower in men compared to women. The decrease in exposure in male patients falls within the comparability bounds of (0.6, 1.6). CDI recurrence rates were the same (17%) in men and women treated with 10 mg/kg bezlotoxumab and the safety profile was consistent between men and women. Thus, the

effect of gender on bezlotoxumab $AUC_{0\text{-}inf}$ is not clinically meaningful and no dose adjustment is warranted.

*2.3.2.6. Race*

The majority of individuals in the population pharmacokinetic analysis were Caucasian (83%), with African-American (AA), and Asian individuals representing 6% and 9% of the analysis population, respectively, and other (including Native American) individuals representing ~2% of the analysis population. Bezlotoxumab $AUC_{0\text{-}inf}$ was 14% lower in AA patients compared to non-AA patients. The slight decrease in exposure in AA patients falls within the comparability bounds of (0.6, 1.6). CDI recurrence rates were similar in Caucasian (17%) patients versus all other races (15%) treated with 10 mg/kg bezlotoxumab. The safety profile following treatment with 10 mg/kg bezlotoxumab was similar in Caucasian patients and all other races.

The effect of Japanese origin on bezlotoxumab PK was also evaluated in the PopPK analysis, which included 80 Japanese individuals from the Phase 1 and Phase 3 trials (5% of the analysis population). Inter-study comparisons of conventional pharmacokinetics following single-dose administration to healthy subjects in Phase 1 studies suggested that $AUC_{0\text{-}inf}$ was 8% lower and $C_{max}$ was 6% lower in Japanese subjects compared to non-Japanese subjects. Bezlotoxumab $AUC_{0\text{-}inf}$ was 15% lower in Japanese patients compared to non-Japanese patients in the Phase 3 patient population, driven largely by lower weight in Japanese patients. This slight decrease in exposure in Japanese patients falls within the clinical experience that defines the comparability bounds of (0.6, 1.6).

Thus, the effect of race on bezlotoxumab $AUC_{0\text{-}inf}$ is not clinically meaningful and no dose adjustment is warranted.

*2.3.2.7. Renal impairment*

Therapeutic mAbs are normally catabolized in a variety of tissues, and are too large (M.W. approximately 150kDa) to pass through the glomerular basement membrane of the kidney. Therefore, no major changes in bezlotoxumab exposure are anticipated in the setting of renal impairment.

Compared to patients with normal renal function, bezlotoxumab $AUC_{0\text{-}inf}$ was 19% higher in patients with mild renal impairment and 16% lower in patients classified as having end-stage renal disease, with $AUC_{0\text{-}inf}$ differences for patients with moderate and severe renal impairment falling between these two effects. The changes in exposure in patients across the levels of renal impairment are limited and fall within the clinical experience that defines the comparability bounds of (0.6, 1.6). CDI recurrence rates were similar regardless of renal function in patients treated with 10 mg/kg bezlotoxumab with serum creatinine concentration (SCr) $\geq$ 1.5 mg/dL (14%) or with SCr < 1.5 mg/dL (17%). Thus, the effect of renal impairment on bezlotoxumab $AUC_{0\text{-}inf}$ is not clinically meaningful and no dose adjustment is warranted.

*2.3.2.8. Hepatic impairment*

No major changes in bezlotoxumab exposure are anticipated in the setting of hepatic impairment. The effect of hepatic impairment on bezlotoxumab PK has not been investigated in clinical studies. The PopPK analysis indicates no effect of hepatic impairment on the pharmacokinetics of bezlotoxumab. Bezlotoxumab exposure was 28% lower in patients with hepatic impairment. The decrease in exposure in patients with hepatic impairment is driven largely by lower albumin concentrations in these patients. The changes in exposure in patients with hepatic impairment are modest and fall within the comparability bounds of (0.6, 1.6). Thus, the effect of hepatic impairment on bezlotoxumab $AUC_{0\text{-}inf}$ is not clinically meaningful and no dose adjustment is warranted.

*2.3.2.9. Pregnancy/Lactation*

Bezlotoxumab has not been studied in pregnant or nursing women. It is not known if bezlotoxumab is excreted in human milk. The Sponsor has recommended that caution should be exercised when bezlotoxumab is administered to a nursing woman and that bezlotoxumab should be used during pregnancy only if clearly needed.

*2.3.2.10. Presence of Comorbid Conditions*

The effect of comorbid conditions, as assessed in the Charlson Comorbidity Index, was evaluated in the PopPK analysis. This covariate was not included in the final PopPK model as it was highly correlated to albumin concentration. The effects of disease severity and patient health on bezlotoxumab $AUC_{0\text{-}inf}$ are not clinically meaningful and no dose adjustment is warranted.

*2.3.2.11. Immunoglobin Levels (endogenous anti-C-difficile toxin B IgG)*

The effect of endogenous anti-*C-difficile* toxin B IgG concentrations (IgG-B) on bezlotoxumab PK was evaluated based on exposures achieved in the Phase 3 population (Study P001 and P002). As endogenous IgG-B concentrations were assessed semi-quantitatively, patients were stratified into three endogenous IgG-B titer concentrations to visualize any trends across titer level. Bezlotoxumab exposures are essentially the same across endogenous IgG-B titer concentrations.

*2.3.3. Immunogenicity*

Overall, the data from CDI patients in the Phase 3 trials indicate a low potential to elicit the formation of treatment emergent anti-drug antibody (ADA) to bezlotoxumab. No treatment emergent ADA to bezlotoxumab has been observed. Pre-treatment and post-dose samples were collected and immunogenicity status was determined for all subjects with at least one post-dose sample assay result. Both Phase 3 trials (Studies P001and P002) and the two later Phase 1 trials which evaluated immunogenicity (Studies P004 and P006) were supported by an ECL-based ADA assay. Please see detail of the assays supporting the immunogenicity assessment for each trial in *Section 2.6*.

*2.3.3.1. What is the incidence (rate) of formation of the anti-drug antibodies (ADA), including the rate of pre-existing antibodies, the rate of ADA formation during and after treatment, time profiles and adequacy of the sampling schedule?*

Bezlotoxumab can interfere with the ADA assay at concentrations above the Drug Tolerance Limit (DTL) (7.5 µg/mL). In the presence of high levels of bezlotoxumab, there is the potential for false negative ADA results. Therefore, samples with a negative test result in the screening ADA assays could only be confirmed as negative when the bezlotoxumab concentration was below the DTL. The immunogenicity status of a patient could then only be confirmed as negative if all pre-treatment and post-dose samples were negative, and if the concentration of bezlotoxumab in the last post-dose sample from each subject was below the DTL.

Two of the Phase 1 trials investigating bezlotoxumab + actoxumab (Studies P006 and P020) were single dose trials, but in one Phase 1 trial (Study P004), a dose was given at both Day 1 and Day 85 to assess immunogenicity following a second dose of bezlotoxumab +actoxumab. The bioanalytical assay for immunogenicity was not specific to assess the presence of ADA against bezlotoxumab in Study P020, and thus the immunogenicity data from Study P020 is excluded. In the Phase 1 studies (Studies P004 and P006), anti-bezlotoxumab antibodies were not observed (**Table 2.3.3.1-1**), indicating lack of immunogenicity of bezlotoxumab. Generally, the bezlotoxumab concentrations at the last post-dose time point of Day 168 in P004 and P006 were below the DTL of 7.5 mcg/mL for the anti-bezlotoxumab antibody assay, suggesting that the negative results were reliable for the majority (>96%) of subjects. The single inconclusive subject terminated early from P004, with the last sample collected 42 days after the administration.

**Table 2.3.3.1-1.** Immunogenicity Status (Presence of Anti-bezlotoxumab Antibodies) in Healthy Subjects Enrolled in Phase 1 Studies Using a validated ECL Assay (Studies P004 and P006)

| Trial | Treatment | Total Treated | Total Evaluable Subjects[†]: | Negative[‡] | Inconclusive[‡] | Dose level with positive subjects | Non-treatment-emergent Positive | Treatment-emergent Positive |
|---|---|---|---|---|---|---|---|---|
| P004 | MK-3415A | 30 | 30 | 29 (96.7%) | 1 (3.3%) | NA | 0 | 0 |
| P006 | MK-3415A | 12 | 12 | 12 (100%) | 0 (0%) | NA | 0 | 0 |
| Summary | | 42 | 42 | 41 (97.6%) | 1 (2.4%) | | 0 | 0 |

[†]: Includes patients with at least one ADA sample available after treatment with MK-3415A.
[‡]: Denominator was total number of evaluable patients.
MK-3415A = actoxumab + bezlotoxumab

The incidences of anti-bezlotoxumab antibodies in the Phase 3 studies are presented in **Table 2.3.3.1-2**. Based on the evaluation of bezlotoxumab concentrations in the last available serum sample, 1013 of the 1414 subjects (71.6%) were reported as negative and 392 of the 1414 subjects (27.7%) were reported as inconclusive, suggesting that the negative results were reliable for the majority of subjects. There were no bezlotoxumab treatment-emergent positive patients, although there were 9 of 1414 patients (0.6%) who had non-treatment-emergent positive samples. Of these nine positive samples, only one was subsequently shown to be positive for neutralizing

antibody (NAb). As no treatment emergent positive patients were observed for bezlotoxumab, no effects on safety or efficacy due to ADA are anticipated.

**Table 2.3.3.1-2** Immunogenicity Status (Anti-bezlotoxumab Antibodies) of Patients with CDI Enrolled in the Phase 3 Trials (Studies P001 and P002).

| Trial | Treatment | Dose (mg/kg) | Total dosed with MK-6072 | Total Evaluable† | Negative‡ | Inconclusive‡ | Non-treatment-emergent Positive‡ | Treatment-emergent Positive‡ |
|---|---|---|---|---|---|---|---|---|
| P001 | MK-6072 | 10 | 390 | 351 | 239 (68.1%) | 112 (31.9%) | 0 (0%) | 0 (0%) |
| | MK-3415A | 10 | 387 | 356 | 237 (66.6%) | 114 (32.0%) | 5 (1.4%) | 0 (%) |
| P002 | MK-6072 | 10 | 396 | 359 | 271 (75.5%) | 88 (24.5%) | 0 (0%) | 0 (0%) |
| | MK-3415A | 10 | 390 | 348 | 266 (76.4%) | 78 (22.4%) | 4 (1.1%) | 0 (%) |
| Phase 3 Summary | | | 1563 | 1414 | 1013 (71.6%) | 392 (27.7%) | 9 (0.6%) | 0 (%) |

†: Includes patients with at least one ADA sample available after treatment with MK-3415A or MK-6072.
‡: Denominator was total number of evaluable patients.
MK-6072 = bezlotoxumab; MK-3415A = actoxumab + bezlotoxumab

*2.3.3.2. Does the immunogenicity affect the PK and/or PD of the therapeutic protein?*

Immunogenicity had no discernable impact on bezlotoxumab PK and PD.

*2.3.3.3. Do the anti-product antibodies have neutralizing activity?*

Only one patient in both Phase 3 studies was subsequently shown to be positive for neutralizing antibody (NAb) which is not treatment-emergent.

*2.3.3.4. What is the impact of anti-product antibodies on clinical efficacy?*

No impact on clinical efficacy is anticipated, since no treatment emergent anti-bezlotoxumab antibody was observed for patients receiving a single dose bezlotoxumab treatment.

*2.3.3.5. What is the impact of anti-product antibodies on clinical safety (e.g. infusion-related reactions, hypersensitivity reactions, cross-reactivity to endogenous counterparts, etc.)?*

Since no treatment emergent positive patients were observed for bezlotoxumab, the impact on anti-product antibodies on clinical safety is expected to be minimal.

## 2.4. Extrinsic Factors

*2.4.1. What extrinsic factors (drugs, herbal products, diet, smoking, and alcohol use) influence dose-exposure and/or -response and what is the impact of any differences in exposure on response? Based upon what is known about exposure-response relationships*

*and their variability, what dosage regimen adjustments, if any, do you recommend for each of these factors?*

As mentioned previously in *Section 2.3*, no extrinsic factors have been shown to influence bezlotoxumab exposure and/or response. Therefore, no dosage adjustments for extrinsic factors are recommended.

*2.4.2. Drug-Drug Interactions*

*2.4.2.1. Is there an in vitro basis to suspect in vivo drug-drug interactions?*

There is no *in vitro* basis to suspect in vivo drug-drug interactions with bezlotoxumab.

*2.4.2.2. Is the drug a substrate of CYP enzymes? Is metabolism influenced by genetics?*

As a monoclonal antibody, bezlotoxumab is expected to be degraded into small peptides and individual amino acids. Therefore, it is not a substrate of CYP enzymes and its metabolism is not influenced by genetics.

*2.4.2.3. Is the drug an inhibitor and/or an inducer of CYP enzymes?*

As a monoclonal antibody, the target of bezlotoxumab is an exogenous toxin and is not a cytokine modulator. Thus, bezlotoxumab is not expected to be an inhibitor and/or an inducer of CYP enzymes.

*2.4.2.4. Is the drug a substrate and/or an inhibitor of P-glycoprotein transport processes?*

As a monoclonal antibody, bezlotoxumab is not expected to be a substrate and/or an inhibitor of P-glycoprotein transport processes.

*2.4.2.5. Are there other metabolic/transporter pathways that may be important?*

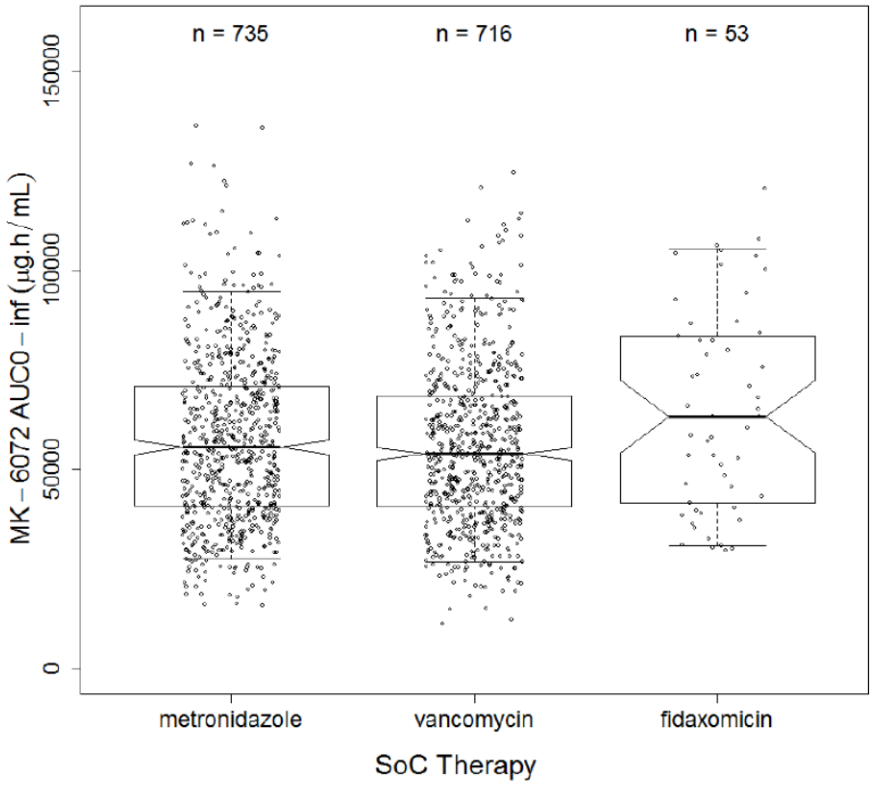
Since bezlotoxumab is a monoclonal antibody, other metabolic/transporter pathways are not expected to be of importance.

*2.4.2.6. Does the label specify co-administration of another drug (e.g., combination therapy in oncology) and, if so, has the interaction potential between these drugs been evaluated?*

The DOSAGE AND ADMINISTRATION section of the proposed labeling recommends that bezlotoxumab should be administered (b) (4) for CDI. In the Phase 3 studies, the standard of care (SoC) antibiotic was oral metronidazole, vancomycin or

fidaxomicin. In addition, subjects receiving vancomycin or fidaxomicin could also receive IV metronidazole. There is no evidence of significant change in bezlotoxumab PK exposure during a course of antibiotic therapy for CDI (**Figure 2.4.2.6-1**).

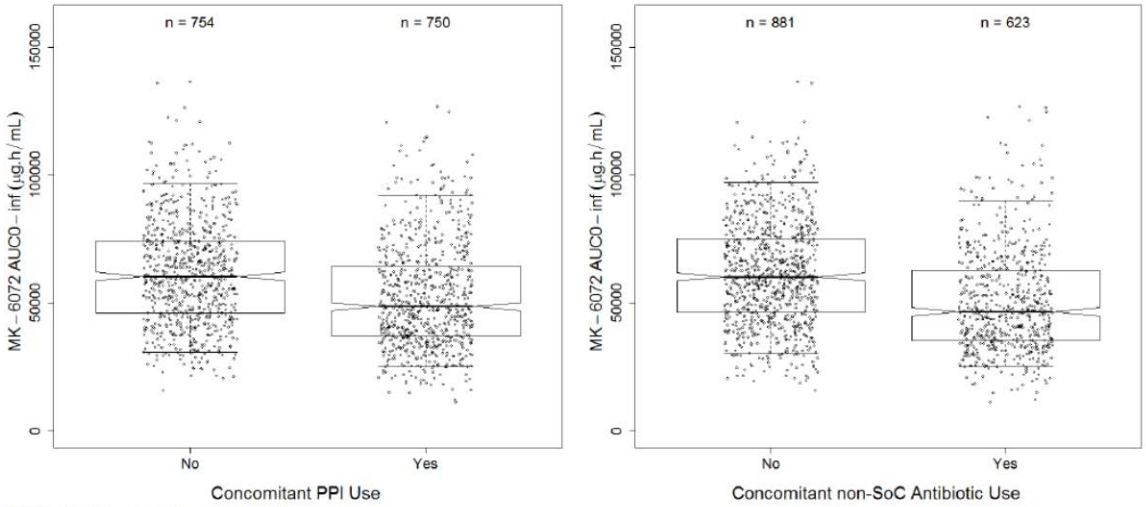
**Figure 2.4.2.6-1** Distribution of Bezlotoxumab $AUC_{0\text{-}inf}$ in Patients across the Standard of Care Therapies Following Administration of 10 mg/kg Bezlotoxumab or 10 mg/kg Actoxumab + Bezlotoxumab




SoC = standard of care, MK-6072 = bezlotoxumab
Note: The boxes represent 25th and 75th percentiles; the line reflects the median; the whiskers are 5th and 95th percentiles.

Concomitant use of non-SoC systemic antibiotics and proton pump inhibitors (PPIs) were also evaluated as potential covariates on the PK of bezlotoxumab in the PopPK analysis. As shown in **Figure 2.4.2.6-2,** mean values of bezlotoxumab exposures were 20% and 16% lower in patients using non-SoC systemic antibiotics or PPIs, respectively, compared to patients not using these concomitant medications. CDI recurrence rates were similar in patients regardless of non-SoC systemic antibiotic or PPI use.

**Figure 2.4.2.6-2** Distribution of Bezlotoxumab $AUC_{0\text{-}inf}$ in Patients With and Without Concomitant Proton Pump Inhibitor (PPI) or non-Standard of Care Systemic Antibiotic Use Following Administration of 10 mg/kg Bezlotoxumab or 10 mg/kg Actoxumab + Bezlotoxumab




MK-6072 = bezlotoxumab
Note: The boxes represent 25th and 75th percentiles; the line reflects the median; the whiskers are 5th and 95th percentiles.

*2.4.2.7. Are there any in vivo drug-drug interaction studies that indicate the exposure alone and/or exposure-response relationships are different when drugs are co-administered?*

No *in vivo* drug-drug interaction studies were conducted. In some of the Phase 1 trials and both Phase 3 trials, actoxumab was co-administered with bezlotoxumab in a combination of actoxumab and bezlotoxumab. Bezlotoxumab pharmacokinetic parameters $AUC_{0\text{-}inf}$ and $C_{max}$ after administration of 10 mg/kg bezlotoxumab alone were similar to those after administration of a combination of actoxumab + bezlotoxumab (10 mg/kg each) in healthy subjects (Study P020). In addition, in the population PK analysis across healthy subjects and patients (n=1587 subjects), co-administration with actoxumab did not have a significant effect on bezlotoxumab pharmacokinetics ($p > 0.05$).

*2.4.2.8. Is there a known mechanistic basis for pharmacodynamic drug-drug interactions, if any?*

No, there is no known mechanistic basis for pharmacodynamic drug-drug interactions.

*2.4.2.8.1. Are there any unresolved questions related to metabolism, active metabolites, metabolic drug interactions, or protein binding?*

There are no unresolved issues related to metabolism, active metabolites, metabolic drug interactions or protein binding for this monoclonal antibody.

## 2.5. General Biopharmaceutics

Not applicable.

## 2.6. Analytical Section

This section summarizes the bioanalytical methods utilized to assess therapeutic protein concentrations. Details for the bioanalytical methodology used to determine bezlotoxumab serum concentrations in Phase 1 clinical trials (Studies P004, P005, and P006) and the pivotal Phase 3 trials (Studies P001 and P002) are presented in the individual study reviews in *Appendix 4.1*.

*2.6.1. What bioanalytical methods are used to assess the therapeutic protein concentrations? Briefly describe the methods and summarize the assay performance.*

In an initial Phase 1 trial (Study P020) and a Phase 2 trial (Study P017), bezlotoxumab concentration measurements and anti-drug antibody (ADA) assessments in serum were performed using a ligand-capture-based enzyme-linked immunosorbent assay (ELISA). This method was not able to distinguish endogenous anti-toxin B antibodies vs. bezlotoxumab. The concentration measurements from the ELISA assays were a combination of endogenous anti-toxin B antibodies and bezlotoxumab. Thus, the bezlotoxumab serum concentration data obtained with this assay are not meaningful and should not be included in the bezlotoxumab PK assessment. Therefore, the detail of this ELISA assay for bezlotoxumab detection in serum was not reviewed.

Subsequent Phase 1 clinical trials (Studies P004, P005, and P006) and the pivotal Phase 3 trials (Studies P001 and P002) employed a conventional sandwich electrochemiluminescence (ECL) immunoassay for the quantitation of the serum concentration of bezlotoxumab, which was specific for bezlotoxumab. Briefly, an internally produced mouse monoclonal anti-idiotype anti-bezlotoxumab antibody (MP770-15B7-5) is biotinylated and used as the capture reagent. It is also labeled with Meso Scale Discovery® SULFO-TAG™ and used as the detection reagent. Calibration standard, quality control (QC), and test samples were analyzed after a minimum required dilution (MRD) of 1:10. ECL response (as relative luminescence units, RLU) from the bound mouse anti-bezlotoxumab SULFO-TAG™ was measured on the Meso Scale Discovery® platform. The bezlotoxumab concentration in test samples was then determined by interpolation using a weighted 5-parameter logistic model algorithm to fit a standard curve that ranged in serum concentrations from 100 (LLOQ) to 6400 (ULOQ) ng/mL. There was no interference due to the presence of actoxumab up to the maximum tested concentration of 400 μg/mL, which approximates the arithmetic mean $C_{max}$ (421 μg/mL) observed following a 20 mg/kg dose of actoxumab, the highest dose tested in clinical development. This ECL immunoassay for bezlotoxumab measurement in serum is acceptable. Assay specifics and performance are summarized in **Table 2.6.1-1**.

**Table 2.6.1-1**. Summary of Validation Parameters and Performance for the ECL Immunoassay for Bezlotoxumab Concentration in Human Serum

| Validation Parameter | Assay Performance |
|---|---|
| ***Parameters established at*** [redacted] (b) (4) ***using automated sample dilutions*** | |
| Assay Range | 100 – 6400 ng/mL expressed in 100% human serum |
| Standard Curve Accuracy | % Bias between -6.07% to 5.85% |
| Standard Curve Precision | % CV ≤ 3.72% |
| QC Accuracy | Intra Assay % Bias between -2.73% to 10.4% |
| | Inter Assay % Bias between -1.36% to 6.80% |
| QC Precision | Intra Assay % CV ≤ 7.58% |
| | Inter Assay % CV ≤ 5.72% |
| Dilutional Linearity | Up to 44000 ng/mL |
| Storage Stability of QC samples and DQC Diluted to MRD | Up to 24 hours at temperature between 2°C to 8°C |
| Automated Dilution Scheme A using Tecan Evo | Intra Assay % Bias between -6.93% to -4.68% |
| | Intra Assay % CV ≤ 5.54% |
| Automated Dilution Scheme B using Tecan Evo | Intra Assay % Bias between -5.60% to -2.39% |
| | Intra Assay % CV ≤ 5.82% |
| Automated Dilution Scheme C using Tecan Evo | Intra Assay % Bias between -4.40% to 4.37% |
| | Intra Assay % CV ≤ 5.99% |
| ***Other parameters previously established at Merck, Union, NJ*** | |
| Sensitivity (LLOQ) | 100 ng/mL expressed in 100% human serum |
| Selectivity | Normal: 5 out of 5 normal serum samples were within acceptance criteria of ±25% of nominal and with a CV ≤20% for QCs at 300 and 5120 ng/mL |
| | CDI: 5 out of 5 disease state samples were within acceptance criteria of ±25% of nominal and with a CV ≤20% for QCs at 300 and 5120 ng/mL |
| Room Temperature QC Stability | Up to 24 hours |
| Freeze-Thaw QC Stability | Up to 6 Freeze/Thaw cycles |
| Long Term QC Stability | Up to 7 days at 4°C |
| | Up to 4 months at -20°C |
| | Up to 12 month at -80°C |
| Long Term Top Standard Storage Stability | Up to 12 month at -80°C |
| Co-dosing Interference | MK-3415† Up to 400 µg/mL, no interference |
| † MK-3415 = actoxumab and MK-6072 = bezlotoxumab | |

LLOQ, lower limit of quantitation

Additionally, a conventional sandwich ECL method for the detection of bezlotoxumab presence in stool was developed, characterized, and used for samples from the clinical trial P002. The ECL-based assay for the detection of bezlotoxumab in stool samples uses the same anti-idiotype antibody as used in the serum assay, coupled to either biotin or SULFO-TAG™ as capture and detection reagents on the Meso Scale Discovery® platform. As a result of the heterogeneity of the

matrix, the assay showed limited matrix selectivity. The assay was therefore developed and characterized as a semi-quantitative assay. The LLOQ for the bezlotoxumab assay in human stool was 160 ng/mL. The assay performance characterization including accuracy, precision, selectivity, and stability is summarized in (**Table 2.6.1-2**)

**Table 2.6.1-2** Summary of Performance Characteristics for the Bezlotoxumab Detection Assay in Human Stool

| **Characterization Parameter** | **Assay Performance** |
|---|---|
| ***Parameters established at*** (b) (4) | |
| Assay Range | 16.0 – 512 ng/mL expressed in 10% stool supernatant |
| Standard Curve Accuracy | % Bias between -3.6 to 5.6% |
| Standard Curve Precision | %CV ≤ 21.6% |
| QC Accuracy | Intra Assay % Bias between -11.4 and 14.7% |
| | Inter Assay % Bias between -26.0 and 26.9% |
| QC Precision | Intra Assay %CV ≤ 6.3% |
| | Inter Assay %CV ≤ 11.1% |
| Sensitivity (LLOQ) | 160 ng/mL expressed in 100% stool |
| Selectivity | Normal population (samples spiked with MK-6072†):<br>Solid stools: 2 out of 10 tested >LLOQ<br>Semi-solid stools: 5 out of 12 tested >LLOQ<br>Liquid stools: 10 out of 10 tested >LLOQ |
| | CDI population (samples spiked with MK-6072†):<br>Semi-solid stools: 7 out of 8 tested >LLOQ<br>Liquid stools: 2 out of 2 tested >LLOQ |
| Long Term QC Stability | Up to 1 month at -20 °C |
| | Up to 12 months at -70 °C |
| † MK-6072 = bezlotoxumab | |

*2.6.1.1. What is the range of the standard curve? How does it relate to the requirements for clinical studies? What curve fitting techniques are used?*

As mentioned previously, the standard curve ranged in serum concentrations from 100 to 6400 ng/mL. The standard curve was a weighted 5-parameter logistic model algorithm with concentration of analyte (X) as the independent variable and absorbance (Y) as the dependent variable:

$$Y = D + (A - D) / (1 + (X / C)^B)^M$$

In this expression, A refers to the "zero" dose response, D refers to the infinite dose response, C refers to the analyte concentration that produces 50% of the maximum response, B is a slope factor related to the shape and steepness of the curve, and M is asymmetry factor. The regression was performed with $1/Y^2$ weighting.

The ranges of standard curves for serum concentration assays are adequate for purposes of determining serum concentrations of bezlotoxumab in the pivotal clinical studies.

*2.6.1.2. What are the lower and upper limits of quantification (LLOQ/ULOQ)?*

The LLOQ and ULOQ are 100 and 6400 ng/mL in 100% serum, respectively.

*2.6.1.3. What are the accuracy, precision, and selectivity at these limits?*

Refer to **Table 2.6.1-1** for the accuracy and precision for each bezlotoxumab assay.

*2.6.1.4. What is the sample stability under the conditions used in the study (long-term, freeze-thaw, sample-handling, sample transport, autosampler)?*

Refer to **Table 2.6.1-1** for bezlotoxumab sample stability under various storage conditions: (1) at room temperature; (2) frozen at -80ºC and thawed 6 times; (3) stored for 7 days at 4ºC; (4) stored for 4 months at -20 ºC; and (5) stored for 12 months at -80ºC.

*2.6.1.5. What is the QC sample plan?*

Bezlotoxumab concentrations in 100% human serum were prepared as QCs: 300 ng/mL (LQC), 800 ng/mL (MQC), 5120 ng/mL (HQC), 44000 ng/mL (DQC1), 600000 ng/mL (DQC2). Bezlotoxumab LQC, MQC and HQC served as the assay control QCs as well as the stability test samples used for refrigerated storage stability. DQC1 was used to perform dilutional linearity to evaluate the alternate method of sample dilution. DQC2 was used to evaluate the performance of the TECAN EVO workstation to perform sample dilutions and the MRD. DQC2 was also used to evaluate refrigerated storage stability. Four out of the six QC samples (low, mid and high QCs x 2 determinations) assayed in replicate on a plate must be within 20% of their nominal values.

*2.6.2. What bioanalytical methods are used to assess the formation of the anti-product antibodies? Briefly describe the methods and summarize the assay performance, including sensitivity, specificity, precision, cut point, interference and matrix, etc.*

*2.6.2.1. What is the performance of the binding assay(s)?*

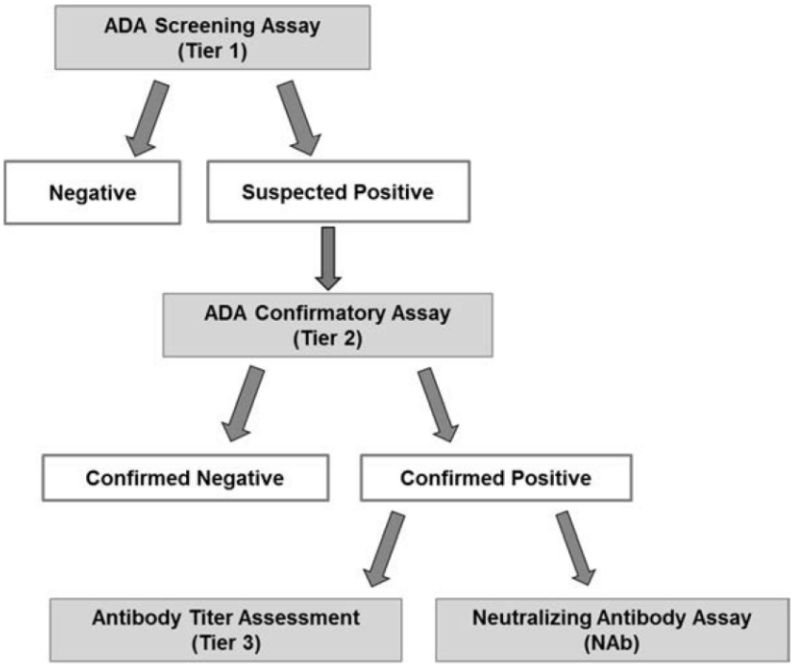
Two Phase 1 trials (Studies P004 and P006) and the two pivotal Phase 3 trials (Studies P001 and P002) included an ECL-based ADA assay that was validated according to current guidance and industry standards (FDA 2009 draft guidance and EMA 2012 guidance for immunogenicity assays). No ADA samples were collected in P005 (Phase 1). Bioanalysis of ADA using the validated ECL assay was carried out using a typical 3-tiered assay approach that consists of:

• Tier 1 ADA screening: to determine if ADA is potentially present (suspected positives), using a screening assay cut point that identifies a target of 5% false positives

• Tier 2 Confirmatory assay: to determine if the signal is specific for the therapeutic protein, using a confirmatory assay cut point that allows ~1% false positives
• Tier 3 Titer: to semi-quantify the ADA response by assessing the titer of ADA in confirmed samples
• Tier 3 NAb assay: Tier 2 confirmed ADA positive samples are also tested in a cell-based NAb assay to assess the ability of ADA to block (neutralize) bezlotoxumab from binding to its in vivo target.

A flowchart of the standard ADA sample analysis is given in (**Figure 2.6.2.1-1**).

**Figure 2.6.2.1-1** ADA Assay Paradigm Details for Tier 1, 2, and 3




ECL-based bridging ADA assay uses biotin- and ruthenium-labeled bezlotoxumab for capture and detection of anti-bezlotoxumab antibodies. Positive and negative controls and test specimens are diluted to an MRD of 50% serum and subsequently co-incubated with biotinylated bezlotoxumab and SULFO-TAG™-labeled bezlotoxumab in individual tubes to create a “master mix” leading to a final solution containing 17% serum. Following incubation, aliquots of the master mix are pipetted onto a blocked Meso Scale Discovery® streptavidin coated standard MA2400 96-well plate. Unbound material is removed by washing the plate, and the ECL response (or relative luminescence units, RLU) from the bound bezlotoxumab SULFO-TAG™ is detected using the Meso Scale Discovery® platform. An increase in the signal response over the cut point indicates the potential presence of bezlotoxumab reactive antibodies. The assay’s cut point is defined as the level of signal response above which a sample is defined to be potentially positive and at or below which it is defined to be negative for the presence of ADA. The assay limit of detection (LOD)/sensitivity was 0.189 ng/mL, established using a hyperimmunized rabbit

polyclonal anti-bezlotoxumab antibody. The cut point of the assay was assessed using serum samples from bezlotoxumab-naïve CDI subjects and was set to target a 5% false positive rate.

The Tier 2 confirmatory assay uses the same approach as the Tier 1 screening assay, except that excess unlabeled bezlotoxumab is added to the specimen at a final concentration of 2.4 μg/mL (expressed in 100% serum). Samples that demonstrate competitive inhibition of the assay's response signal greater than the confirmatory cut point after addition of bezlotoxumab are classified as being positive for the presence of anti-bezlotoxumab antibodies. The confirmatory cut point was assessed using serum samples from bezlotoxumab-naïve CDI patients and allowed a 1% false positive rate.

For Tier 3 analysis, samples are diluted serially in pooled normal human serum prior to analysis in the screening assay for the determination of the titer of an ADA-confirmed sample. Only Tier 2-confirmed ADA positive samples are reported with a titer value. The performance of the validated method is summarized in **Table 2.6.2-1**. Please also refer to Dr. William Hallet's review (Office of Biotechnology Products) for more details.

**Table 2.6.2-1**.Summary of Assay Validation Parameters and Performance for the Second Generation Bezlotoxumab ADA Assay

| Validation Parameter | Assay Performance |
|---|---|
| ***Parameters established*** (b) (4) | |
| Cut Point Factor (Normalization Correction Factor, NCF)<br>Cut point = Negative Control response + NCF | Normal: 5.95 ECL counts |
| | CDI: 1.93 ECL counts |
| Specificity (Confirmatory) Cut Point<br>Specificity cut point =<br>(1-(inhibited/uninhibited)) x100% | Normal: 39.5% |
| | CDI: 34.6% |
| Precision<br>Screening Assay | Intra-assay: ≤ 5.66%<br>(Target: ≤ 30%) |
| | Inter-assay: ≤ 18.1%<br>(Target: ≤ 30%) |
| Precision<br>Titer Assay | Intra-assay: ≤ 14.1%<br>(Target: ≤ 50%) |
| | Inter-assay: ≤ 18.2%<br>(Target: ≤ 50%) |
| Sensitivity<br>(anti-CDR enriched Rabbit polyclonal anti-MK-6072† antibody) | 0.189 ng/mL |
| Drug Tolerance | MK-6072†: at 500 ng/mL of anti-MK-6072, tolerance up to 7.5 µg/mL MK-6072 |
| | MK-3415†: at 0.823 ng/mL anti-MK-6072, tolerance of at least 10 µg/mL MK-3415 |
| ***Other parameters previously established at Merck, Union, NJ*** | |
| Robustness<br>Screening Assay | Short Incubation Time: 1 hr/0.5 hr met acceptance criteria (HPC>LPC>Cut Point) |
| | Long Incubation Time: 2.5 hr/1.5 hr met acceptance criteria (HPC>LPC>Cut Point) |
| Matrix Selectivity | Normal: 5/5 individuals spiked at 2 ng/mL and 200 ng/mL ADA met acceptance criteria (HPC>LPC>Cut Point) and showed <25% Bias |
| | CDI: 5/5 individuals spiked at 2 ng/mL and 200 ng/mL ADA met acceptance criteria (HPC>LPC>Cut Point) and 4/5 individuals showed <25% Bias |
| Specificity/Cross Reactivity | Anti-SCH717454‡, Anti-SCH900222‡, Anti-SCH900475‡ and Anti-MK-3415 were spiked at 2.00 ng/mL and 200 ng/mL. At 200 ng/mL Anti-MK-3415 generated a signal above the cut point. All other samples were below the cut point. |
| Positive Control Storage Stability | Overnight at Room Temperature |
| | Up to 7 days in a refrigerator set to 4°C |
| | Up to 6 months in a freezer set to -20°C |
| | Up to 12 months in a freezer set to -80°C |
| Freeze/Thaw Positive Control Stability | Up to 5 Freeze/Thaw cycles |

† MK-3415 = actoxumab and MK-6072 = bezlotoxumab

‡ SCH717454 = humanized monoclonal antibody against insulin-like growth factor receptor (IGFR)

‡ SCH900222 = humanized monoclonal antibody against human Interleukin-23(IL-23)

‡ SCH900475 = humanized monoclonal antibody against human Programmed cell death protein 1 (PD-1)

*2.6.2.2. What is the performance of the neutralizing assay(s)?*

All samples that were positive in the anti-bezlotoxumab ADA confirmation assay were analyzed in the cell-based neutralizing ADA (NAb) screening and confirmatory assay. A cell-based NAb assay in human serum was developed with the toxin B sensitive cell line IMR-90. The bezlotoxumab NAb assay follows a cell viability assay format. If NAb are present in a human serum sample, bezlotoxumab's ability to bind toxin B will be neutralized and an increase in cytotoxicity will occur. A decrease of cell proliferation is indicative of the presence of neutralizing ADA in a human serum sample. The assay LOD/sensitivity was 10.6 μg/mL established using a hyperimmunized rabbit polyclonal anti-bezlotoxumab antibody. Please also refer to Dr. William Hallet's review (Office of Biotechnology Products) for more details.

## 3. APPENDICES

### 3.1. Individual Study Reviews

**STUDY NO.: PROTOCOL 004-00 (P004)**
**Title: A Phase 1 Study to Evaluate the Immunogenicity of MK-3415A**

**Date(s):** 12-Apr-2013 – 13-Feb-2014
**Investigator(s):** Stephen Youngberg, MD; Charles S. Tomek, MD. Celerion, Lincoln, Nebraska 68502
**Study Center(s):** Celerion, 621 Rose Street, Lincoln, Nebraska 68502
**Analytical Site(s):** (b) (4)

*Note: This study evaluated MK-3415A, a combination of monoclonal antibodies MK-3415 and MK-6072, but the current NDA submission intends to register MK-6072 only. Thus, the current Clinical Pharmacology review will focus on the study result of MK-6072.*

**OBJECTIVE(S):**
**Primary:** To evaluate the immunogenicity of 2 infusions of MK-3415A.
**Secondary:**
1: To evaluate the safety and tolerability of 2 infusions of MK-3415A.
2: To evaluate the pharmacokinetics of 2 infusions of MK-3415A.

**METHODS**
**Study Design:** This was an open-label, multiple-dose study. Thirty (30) healthy adult male and female subjects were enrolled in order to achieve at least 24 evaluable subjects. If more than 6 subjects had discontinued from the study for any reason prior to the administration of the second dose of study drug, additional subjects were to have been enrolled to achieve at least 24 evaluable subjects. On Day 1, subjects received an IV infusion of MK-3415A (10 mg/kg of each mAb, MK-3415 and MK-6072) over 1 hour, followed by pharmacokinetic and immunogenicity sampling for 84 days at specified times. On Day 85, subjects received a second IV infusion of MK-3415A (10 mg/kg dose of each mAb, MK-3415 and MK-6072) over 1 hour, followed by pharmacokinetic and immunogenicity sampling for 168 days at specified times. Safety was monitored throughout the study by repeated assessment of adverse and serious adverse experiences, physical examinations, vital signs, electrocardiograms (ECGs), infusion site reactions, and laboratory evaluations. The schedule of clinical observations and laboratory measurements is shown in **Appendix 1**. The product description and manufacturing lot numbers are provided in **Table 1**.

**Diagnosis/inclusion criteria**: Adult healthy male (weighing ≥ 52 kg) or female subjects (weighing ≥ 45 kg), between the ages of 19 - 55 (inclusive), with a body mass index (BMI) of 18.5 - 32.0 kg/$m^2$ (inclusive) were enrolled in this study.

**Table 1**. Clinical Supplies Dispensed to Subjects

| Drug | Potency | Formulation No. | Dosage Form | Control No. | Assay Potency (ELISA) (%) | Site of Manufacture |
|---|---|---|---|---|---|---|
| MK-3415 | 25 mg/mL | WL00048371 | IV solution | WL00051348/ WL00053103 | 122.6 | Merck Sharp & Dohme Corp. (West Point, PA) |
| MK-6072 | 25 mg/mL | WL00048262/ WL00051105 | IV solution | WL00051349/ WL00053104 | 91.6/91.5 | Merck Sharp & Dohme Corp. (West Point, PA) |
| 0.9% Sodium Chloride† | Not Applicable | Not Applicable | IV solution | Not applicable | Not Applicable | Not Applicable |

†0.9% sodium chloride manufactured by (b) (4), was purchased by the Investigator. The lot number was J3C683; expiration date Sep-2015.

Immunogenicity: Blood samples for determination of immunogenicity (measured as anti-MK-6072 antibodies) were collected at predose of Day 1 and at specified time points on Days 43, 85, 127, 169, and 253.

Pharmacokinetics: Blood samples for determination of MK-6072 serum concentrations were collected at predose and specified time points over 8 hours on Day 1 and on Days 2, 8, and 43 postdose following the first infusion. Blood samples for MK-6072 following the second infusion, administered on Day 85, were collected at predose and specified time points over 8 hours on Day 85 and on Days 86, 92, 127, 169, and 253 postdose. The serum pharmacokinetic parameters ($AUC_{0-\infty}$, $AUC_{0-84}$ days [calculated from predose Day 1 through time corresponding to 0 Hour on Day 85 relative to start of each of the 2 infusions], $AUC_{0-last}$ [reported following second infusion only], $AUC\%_{extrap}$, $C_{max}$, $T_{max}$, $V_{ss}$, CL, and the apparent terminal $t_{½}$) of MK-6072 were calculated following each infusion. Additionally, for all subjects, a blood sample was also collected (predose on Day 1) and archived for future biomedical research (FBR).

Safety: The safety and tolerability of 2 infusions of MK-3415A were monitored through physical examination, vital signs, ECGs, adverse events, infusion site reactions, and clinical laboratory safety tests (hematology, serum chemistry, and urinalysis).

**Analytical Methods:**

Anti-MK-6072 Antibodies in Serum for Immunogenicity

Serum samples collected for immunogenicity (as assayed for anti-MK-6072 antibodies) were analyzed by (b) (4). A validated bridging electrochemiluminescent (ECL) immunoassay was used for the detection of anti-MK-6072 antibodies in human serum. Bioanalysis of MK-6072 anti-drug antibody (ADA) was carried out using the standard 3-tiered assay approach that consisted of screening (Tier 1), confirmation (Tier 2) and antibody titer assessment (Tier 3). Only Tier 2 confirmed ADA positive samples were reported with a titer value. Details of the analytical method to measure the serum anti-MK-6072 antibody are provided in **Appendix 2**.

Detection of Neutralizing Antibodies to MK-6072

The neutralizing ADA assay was based on the ability of ADA to block (neutralize) the critical first step in the pharmacological action of MK-6072, which is binding to *C. difficile* toxin B. For neutralizing antibody (Nab) confirmation, Protein G was used to deplete IgG in serum samples, thereby leading to a reversal of the signal in the NAb assay. If the NAb signal can be reversed by Protein G, this provided supportive evidence that the substance generating the signal was IgG in nature. The NAb bioassay utilized the IMR 90 cell line to detect antibodies present in human serum samples which may neutralize the biological activity of MK-6072.

Since none of the subjects had samples which tested positive for MK-6072 ADA, no cell-based neutralizing ADA assay was performed for NAbs to MK-6072.

MK-6072 in Serum for Pharmacokinetics

Serum samples collected for MK-6072 were analyzed by (b) (4) The analytical method used was an ECL assay. The lower limit of quantitation (LLOQ) for MK-6072 was 100 ng/mL and the analytical range was 10.0 to 640 ng/mL using a 1:10 minimum dilution. Details of the analytical method of the serum MK-6072 concentrations are shown in **Appendix 3**.

*Reviewer comments:*

*Overall, the bio-analytical methods to assess MK-6072 (Bezlotoxumab) and anti-MK-6072 antibody concentrations in human serum are acceptable.*

**RESULTS**

**Study Population:** Thirty (30) healthy male and female subjects were enrolled into the study and 29 subjects completed the study per protocol. One (1) subject (AN 0023) did not return for the second infusion (Day 85) and was lost to follow-up. Data from all 30 subjects were included in the evaluation of immunogenicity, safety, and pharmacokinetics following the first infusion of MK-3415A, as available. Pharmacokinetic analyses for the missing subject following the first infusion were based on available data through 1008 hours post Day 1 infusion. Twenty-nine (29) subjects were included in the evaluation of immunogenicity, safety and pharmacokinetics following the second infusion of MK-3415A. Data from all 30 subjects were included in the statistical analysis of parameters $AUC_{0-\infty}$, $AUC_{0-84}$ days, $C_{max}$, CL, and Vss. The demographic information for the 30 subjects entered into the study is provided **Table 1**.

**Table 1**. Summary of Demographic Data

| AN | Gender | Race | Ethnicity | Age (yr) | Height (cm) | Weight (kg) | Body Mass Index ($kg/m^2$) |
|---|---|---|---|---|---|---|---|
| 0001 | Male | White | Not Hispanic or Latino | 27 | 187.0 | 106.4 | 30.37 |
| 0002 | Male | White | Not Hispanic or Latino | 19 | 181.0 | 79.8 | 24.27 |
| 0003 | Male | White | Not Hispanic or Latino | 45 | 169.0 | 70.9 | 24.68 |
| 0004 | Female | White | Not Hispanic or Latino | 50 | 162.0 | 59.8 | 22.93 |
| 0005 | Female | White | Not Hispanic or Latino | 49 | 175.0 | 64.5 | 21.07 |
| 0006 | Female | Black or African American | Not Hispanic or Latino | 50 | 156.0 | 68.4 | 28.12 |
| 0007 | Female | White | Not Hispanic or Latino | 46 | 164.0 | 57.8 | 21.40 |
| 0008 | Female | White | Not Hispanic or Latino | 37 | 165.0 | 53.0 | 19.51 |
| 0009 | Female | White | Not Hispanic or Latino | 40 | 174.0 | 78.3 | 25.95 |
| 0010 | Male | White | Not Hispanic or Latino | 32 | 168.0 | 66.8 | 23.56 |
| 0011 | Female | White | Not Hispanic or Latino | 31 | 161.0 | 74.7 | 28.72 |
| 0012 | Male | Black or African American | Not Hispanic or Latino | 33 | 170.0 | 69.0 | 23.96 |
| 0013 | Male | White | Not Hispanic or Latino | 28 | 174.0 | 96.8 | 31.99 |
| 0014 | Female | White | Not Hispanic or Latino | 44 | 175.0 | 71.9 | 23.47 |
| 0015 | Female | Black or African American | Not Hispanic or Latino | 37 | 160.0 | 77.8 | 30.38 |
| 0016 | Female | White | Not Hispanic or Latino | 51 | 151.0 | 66.2 | 28.88 |
| 0017 | Male | White | Not Hispanic or Latino | 28 | 179.0 | 68.3 | 21.23 |
| 0018 | Female | White | Not Hispanic or Latino | 29 | 171.0 | 78.6 | 26.81 |
| 0019 | Male | White | Not Hispanic or Latino | 37 | 180.0 | 85.8 | 26.60 |
| 0020 | Male | White | Not Hispanic or Latino | 20 | 175.0 | 69.2 | 22.65 |
| 0021 | Female | White | Not Hispanic or Latino | 34 | 166.0 | 68.9 | 25.03 |
| 0022 | Female | White | Not Hispanic or Latino | 30 | 167.0 | 57.6 | 20.73 |
| 0023 | Male | White | Not Hispanic or Latino | 20 | 186.0 | 89.4 | 25.80 |
| 0024 | Male | White | Not Hispanic or Latino | 24 | 178.0 | 82.0 | 25.79 |
| 0025 | Female | White | Not Hispanic or Latino | 36 | 164.0 | 65.0 | 24.31 |
| 0026 | Male | White | Not Hispanic or Latino | 28 | 178.0 | 74.5 | 23.64 |
| 0027 | Female | White, Black, or African American | Not Hispanic or Latino | 22 | 169.0 | 58.9 | 20.51 |
| 0028 | Male | White | Hispanic or Latino | 23 | 165.0 | 75.1 | 27.73 |
| 0029 | Male | White | Not Hispanic or Latino | 55 | 185.0 | 92.0 | 26.76 |
| 0030 | Male | White | Not Hispanic or Latino | 52 | 173.0 | 77.7 | 25.83 |
| **Study Summary** | | | | | | | |
| N: | | | | 30 | 30 | 30 | 30 |
| Range: | | | | 19 to 55 | 151.0 to 187.0 | 53.0 to 106.4 | 19.51 to 31.99 |
| Arithmetic Mean: | | | | 35 | 170.9 | 73.5 | 25.09 |
| Female N: | | | | 15 | 15 | 15 | 15 |
| Female Range: | | | | 22 to 51 | 151.0 to 175.0 | 53.0 to 78.6 | 19.51 to 30.38 |
| Female Arithmetic Mean: | | | | 39 | 165.3 | 66.8 | 24.52 |
| Male N: | | | | 15 | 15 | 15 | 15 |
| Male Range: | | | | 19 to 55 | 165.0 to 187.0 | 66.8 to 106.4 | 21.23 to 31.99 |
| Male Arithmetic Mean: | | | | 31 | 176.5 | 80.2 | 25.66 |

AN = Allocation number.

**Immunogenicity**

ADA samples for MK-6072 were available from 30 subjects. No treatment-emergent positive subjects were observed in this study. Three (3) subjects (AN 0011, AN 0019, and AN 0022) had pre-treatment samples which were confirmed positive in the assay for MK 3415 ADA and were reported as non-treatment emergent. These 3 samples were also tested in the NAb assay and found to be negative for NAb. The remaining subjects were classified as negative (n = 26) or inconclusive (n = 1, early termination).

None of the subjects had samples which tested positive in the assay for MK-6072 ADA. These subjects were reported as negative for MK-6072 ADA (n = 29) or inconclusive (n = 1, early termination). After treatment with 10 mg/kg MK-3415A and washout for about 6 months, the

drug concentration in the last postdose sample was below the drug tolerance level for both compounds in 96.7% of the subjects.

**Table 2. Summary of Immunogenicity Results**

| | Number of Subjects[†] (% of Total Number of Subjects) | | Immunogenicity Status | | | | |
|---|---|---|---|---|---|---|---|
| | Total | With Last Sample Below DTL[‡] | Negative | Inconclusive | Treatment Emergent Positive | Non-Treatment Emergent Positive | NAb Positive |
| MK-3415 | 30 | 29 (96.7%) | 26 (86.7%) | 1 (3.3%) | 0 | 3 (10.0%) | 0 |
| MK-6072 | 30 | 29 (96.7%) | 29 (96.7%) | 1 (3.3%) | 0 | 0 | 0 |

[†]Subjects with appropriate samples had data from at least 1 sample collected after dosing with MK-3415A.
[‡]DTL: Drug Tolerance Level of ADA assay (15 µg/mL MK-3415, 7.5 µg/mL MK-6072).
Treatment-emergent positive: Pre-treatment sample was negative and at least 1 postdose sample was positive in the confirmatory assay for antibodies against MK-3415 or MK-6072 (treatment-induced positive. OR Pre-treatment and postdose samples were both positive in the confirmatory assay for antibodies against MK-3415 or MK-6072 and the titer increased postdose (treatment boosted positive).
Non-treatment-emergent positive: Pre-treatment sample was positive and postdose sample was negative in the confirmatory assay for antibodies against MK-3415 or MK-6072. OR Pre-treatment and postdose samples were positive in the confirmatory assay for antibodies against MK-3415 or MK-6072 with no increase in titer postdose.
NAb: Neutralizing Antibody.

*Reviewer Comment:*
*Immunogenicity test was positive or inclusive for MK-3415 or MK-6072 in some of healthy subjects, although these subjects were not exposed to MK-3415 or MK-6072. This observation may suggest that the specificity of the immunogenicity assay can be influenced by certain unspecific binding.*

**Pharmacokinetics:**
The MK-6072 $AUC_{0-\infty}$, $AUC_{0-84}$ days, and $C_{max}$ geometric mean ratio (90% CI) values for second infusion/first infusion were 1.07 (1.04, 1.10), 1.05 (1.03, 1.08), and 1.04 (1.02, 1.07), respectively, indicating minimal accumulation in MK-6072 exposure when the second dose is administered 84 days after the first dose. The arithmetic mean serum concentration-time profiles of MK-6072 following administration of MK-3415A as a single 1-hour infusion to healthy adult subjects are presented below in **Figures 1**. **Table 3** presents the statistical summaries of MK-3415 and MK-6072 serum pharmacokinetics in healthy adult subjects following administration of MK-3415A (10 mg/kg of each mAb, MK-3415 and MK-6072) as 2 consecutive single 1-hour infusions, separated by 84 days. Overall, MK-6072 exhibited similar pharmacokinetics following the second infusion relative to the first infusion of 10 mg/kg of MK-3415A.

**Figure 1.** Arithmetic Mean (± SD) Serum Concentration-Time Profiles of MK-6072 Following the First and Second Infusion of MK-3415A in Healthy Adult Subjects (N = 30 First Infusion/N = 29 Second Infusion) (Left, First 24 Hours Postdose; Right, Entire Sampling Interval)

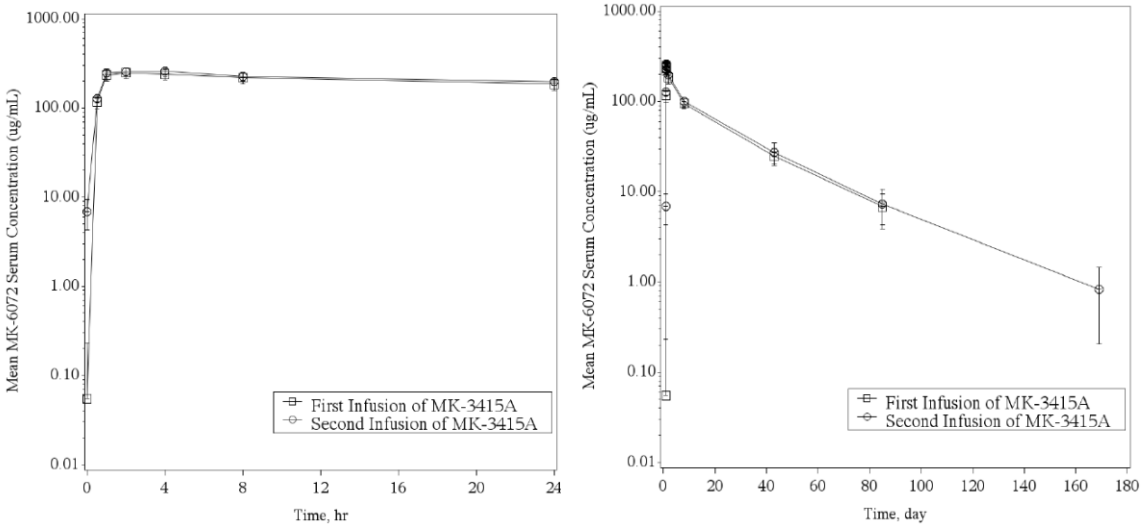



**Table 3**. Statistical Comparison of Serum Pharmacokinetics of MK-6072 Following the Administration of 10 mg/kg MK-3415A as a 1-Hour Infusion in Healthy Adult Subjects

| | | First Infusion of MK-3415A | | | Second Infusion of MK-3415A | | | Second Infusion/ First Infusion | | Pseudo Within Subject %CV[‖] |
|---|---|---|---|---|---|---|---|---|---|---|
| **Monoclonal Antibody** | **Pharmacokinetic Parameter** | **N** | **GM** | **95% CI** | **N[††]** | **GM** | **95% CI** | **GMR** | **90% CI** | |
| MK-6072 | AUC0-∞[†] (µg/mL•hr) | 30 | 85700 | (80500, 91200) | 29 | 91300 | (84700, 98400) | 1.07 | (1.04, 1.10) | 6.249 |
| | AUC0-84 days[†] (µg/mL•hr) | 30 | 81200 | (76700, 85900) | 29 | 85400 | (80100, 91200) | 1.05 | (1.03, 1.08) | 5.772 |
| | Cmax[†] (µg/mL) | 30 | 250 | (238, 264) | 29 | 261 | (251, 271) | 1.04 | (1.02, 1.07) | 5.409 |
| | Vss[†] (mL) | 30 | 5381.65 | (5052.29, 5732.48) | 29 | 5312.16 | (4940.45, 5711.83) | | | |
| | CL[†] (mL/hr) | 30 | 8.57 | (7.89, 9.31) | 29 | 8.05 | (7.36, 8.81) | | | |
| | Tmax[‡] (hr) | 30 | 2.00 | (1.00, 4.08) | 29 | 2.01 | (1.00, 4.02) | | | |
| | Apparent terminal $t_{1/2}$[¶] (hr) | 30 | 474.46 | 15.42 | 29 | 541.80 | 21.27 | | | |

First Infusion (Day 1): Single IV dose of MK-3415A (10 mg/kg of each mAb, MK-3415 and MK-6072) as a 1-hour infusion.
Second Infusion (Day 85): Single IV dose of MK-3415A (10 mg/kg of each mAb, MK-3415 and MK-6072) as a 1-hour infusion.
[†]Back-transformed least-squares mean and confidence interval from linear mixed-effects model performed on natural log-transformed values.
[‖]Pseudo within-subject %CV = 100*Sqrt[($\sigma^2_A$ + $\sigma^2_B$ - 2*$\sigma_{AB}$)/2], where $\sigma^2_A$ and $\sigma^2_B$ are the estimated variances on the log scale for the 2 treatment infusions, and $\sigma_{AB}$ is the corresponding estimated covariance, each obtained from the linear mixed-effects model.
[‡]Median (min, max) reported for Tmax.
[¶]Geometric mean and percent CV reported for apparent terminal $t_{1/2}$.
[††]Subject AN 0023 was lost to follow-up and did not receive the second infusion. Subject AN 0023's AUC0-84 days value was excluded from statistical analysis for first infusion, as subject had no blood draw on Day 84 (his exposure represents data through Day 43 only) following first infusion.
GM = Geometric least-square mean; GMR = Geometric least-square mean ratio; CI = Confidence interval
AUC0-84 days was added as an additional endpoint as the dosing interval was 84 days.

*Reviewer Comment:*
*The PK results of MK-6072 in the present study are consistent with those in the other Phase 1 studies (Studies P020, P005 and P006).*

**Safety:**
Two (2) sequential infusions of MK-3415A were generally well tolerated in the healthy adult males and females in this study. Eighteen (18, 60%) subjects reported a total of 63 post-treatment adverse experiences (22 following first infusion and 41 following second infusion of MK-3415A). The most common adverse experiences reported were headache (30%), oropharyngeal pain (13%) and rhinorrhea (13%). All other adverse experiences were reported by 1 or 2 subjects each overall. Only 1 adverse experience (infusion site extravasation following second infusion) was considered related to the study drug by the Investigator. There were no serious adverse experiences, events of clinical interest, adverse experiences of special interest, pregnancies, deaths, or subject discontinuations due to adverse experiences reported during the study. One (1) subject did not return for the second infusion (Day 85) and was lost to follow-up. No clinically meaningful relationships were observed for differences between clinical laboratory values, vital signs, or ECG safety parameters as a function of treatment.

**SPONSOR'S CONCLUSIONS:**
Following dosing with MK-3415A, no ADAs to MK-6072 were observed. All subjects that washed out MK-3415A for 6 months reduced MK-6072 concentrations low enough to be reported as negative. Overall, the data indicate a low potential of MK-3415A to elicit the formation of anti-drug antibodies.

Two (2) sequential infusions of MK-3415A were generally well tolerated in healthy adult males and females. The serum pharmacokinetics of MK-6072 following 2 sequential 1-hour infusions of 10 mg/kg MK-3415A separated by 84 days were similar ($AUC_{0-\infty}$, GMRs [90% CI] of 1.14 [1.10, 1.19] and 1.07 [1.04, 1.10], respectively), indicating minimal accumulation in exposure of each analyte during the second dose, and no significant changes in pharmacokinetics that often accompany ADA formation.

**REVIEWER ASSESSMENT:**
Following a single dose of 10 mg/kg of MK-3415A, no ADA to MK-6072 was observed and NAb test was negative for MK-6072. The pharmacokinetics of MK-6072 after 1 hour infusion in this study are similar to those in other Phase 1 studies (Studies P005 and P006), and the safety profile appears acceptable. We concur with the Sponsor's conclusion that a low potential of MK-3415A is expected to elicit the formation of ADA against MK-6072.

## Appendix 1. Study Procedures

### Table A1-1. Study Flow Chart

| Study Procedures[a] | | 1st Infusion | | | | | | | | | | 2nd Infusion | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Days → | Screening[b] | -1[c] | 1 | | | | | | 2 | 8 | 43 | 84[c] | 85 | | | | | | 86 | 92 | 127 | 169 | 253 |
| Hours → | | (C-I) | 0 | 0.5 | 1 | 2 | 4 | 8 | | | | (C-I) | 0 | 0.5 | 1 | 2 | 4 | 8 | | | | | |
| **Administrative Procedures** | | | | | | | | | | | | | | | | | | | | | | | |
| Informed Consent | x | | | | | | | | | | | | | | | | | | | | | | |
| Informed Consent for Future Biomedical Research | x | | | | | | | | | | | | | | | | | | | | | | |
| Inclusion/Exclusion Criteria | x | x | | | | | | | | | | x | | | | | | | | | | | |
| Medical History | x | | | | | | | | | | | | | | | | | | | | | | |
| **Safety Evaluations** | | | | | | | | | | | | | | | | | | | | | | | |
| Physical Examination[d] | x | x | | | | | | | | | | x | | | | | | | | | | | x[f] |
| Height | x | | | | | | | | | | | | | | | | | | | | | | |
| Weight | x | x | | | | | | | | | | x | | | | | | | | | | | |
| 12-Lead Electrocardiogram | x | x | | | | | | | | | | x | | | | | | | | | | | x[f] |
| Vital Signs (body temperature, respiratory rate, blood pressure and heart rate ) | x | x | x[e] | | x | x | x | | x | x | x | x | x[e] | | x | x | x | | x | x | x | x | x[f] |
| Hematology, Chemistry, and Urinalysis[g] | x | x | | | | | | | x | | x | x | | | | | | | x | | x | x | x[f] |
| Serum Pregnancy Test (female subjects only) | x | x | | | | | | | | | | x | | | | | | | | | | | |
| Serum FSH (postmenopausal females only) | x | | | | | | | | | | | | | | | | | | | | | | |
| Urine Drug and Alcohol Screen | x | x | | | | | | | | | | x | | | | | | | | | | | |
| HIV/Hepatitis Screen | x | | | | | | | | | | | | | | | | | | | | | | |
| Adverse Events Monitoring | <----------x----------> | | | | | | | | | | | | | | | | | | | | | | |
| Concomitant Medication Monitoring | <----------x----------> | | | | | | | | | | | | | | | | | | | | | | |
| Infusion Site Reaction Monitoring | | | <-----x-----> | | | | | | | | | | <-----x-----> | | | | | | | | | | |
| **Study Drug Administration / Pharmacokinetics** | | | | | | | | | | | | | | | | | | | | | | | |
| MK-3415A Administration (1-hour infusion) | | | <-----x-----> | | | | | | | | | | <-----x-----> | | | | | | | | | | |
| Blood for MK-3415 & MK-6072 Pharmacokinetics | | | x[e] | x | x[h] | x | x | x | x | x | x | | x[e] | x | x[h] | x | x | x | x | x | x | x | x |
| Blood for MK-3415 & MK-6072 Immunogenicity | | | x[e] | | | | | | | | x | | x[e] | | | | | | | | x | x | x |
| **Other Procedures** | | | | | | | | | | | | | | | | | | | | | | | |
| Blood for Future Biomedical Research[i] | | | x | | | | | | | | | | | | | | | | | | | | |
| Confinement in the CRU | | <----------x----------> | | | | | | | | | | <----------x----------> | | | | | | | | | | | |
| Return Visits[j] | | | | | | | | | | x | x | | | | | | | | | x | x | x | x |

a. For details on Procedures, refer to Section 10 of the protocol and/or corresponding appendices.
b. Within 28 days of the first study drug administration.
c. Subjects will be admitted to the CRU at least 10 hours prior to dosing. If necessary, the second dosing on Day 85 can be scheduled within ± 1 day of the scheduled time.
d. A full physical examination will be performed at screening. A symptom driven physical examination may be performed at check-in, and at the end of study at the Investigator's discretion.
e. To be performed before dosing.
f. To be performed at the end of study or prior to early termination. Following early termination, the subject will be asked to return to the clinic or be contacted, if the Investigator's deems necessary, for a follow-up (approximately 14 days after early termination), to determine if any adverse events have occurred since the last visit.
g. Samples for serum chemistry will be obtained following a fast of approximately 8 hours; however, in case of dropout or rechecks, subjects may not have fasted for 8 hours before the serum chemistry sample is taken.
h. To be obtained immediately after the end of infusion.
i. Informed consent for future biomedical research samples must be obtained before the DNA sample. DNA sample for analysis should be obtained predose, on Day 1 (or with the next scheduled blood draw), as the last sample drawn, or at a later date as soon as the informed consent is obtained.
j. Return visits can be scheduled within ± 3 days of the scheduled time on Days 8, 43, 92, 127, 169, and 253.

Abbreviations: C-I = Check-in, CRU = Clinical research unit, FSH = Follicle-stimulating hormone, HIV = Human immunodeficiency virus

## Table A1-2 Study Flow Chart Cohort 2

| | | Study Day | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Pre-study[1] | -1 | 1 | | | | | | | | | | | 8 | Post-Study[1] |
| | | | | | Hours Post Initiation of Infusion | | | | | | | | | | |
| | | | Pre-dose | 0 | 0.5 | 1 | 2 | 4 | 6 | 8 | 12 | 16 | 24 | 168 | 504 |
| Study Procedure | | | | | | | | | | | | | | | |
| Study informed consent | X | | | | | | | | | | | | | | |
| Medical History/Assessment of Inclusion/Exclusion Criteria | X | | | | | | | | | | | | | | |
| HIV/Hepatitis screen(per site SOP) | X | | | | | | | | | | | | | | |
| Drug and alcohol screen (per site SOP) | X | X | | | | | | | | | | | | | |
| Genetic Informed Consent | X | | | | | | | | | | | | | | |
| Genetic Sample Collection | | | X | | | | | | | | | | | | |
| Safety evaluations | | | | | | | | | | | | | | | |
| Physical examination (PE)[2] | X | | X | | | | | | | | | | | | X |
| Weight[2] | X | X | | | | | | | | | | | | | X |
| Height | X | | | | | | | | | | | | | | |
| 12-lead electrocardiogram (ECG)[3] | X | | X | | | X | X | | | X | X | | X | | X |
| Semi recumbent vital signs (HR, BP, RR, oral T)[4] | X | | X | | X | X | X | X | | X | X | X | X | | X |
| Orthostatic vital signs (HR, BP)[4] | | | X | | | X | | | | | | | | | |
| Laboratory safety tests (serum chemistry, CBC, urinalysis)[5] | X | | X | | | | | | | | | | X | | X |
| FSH, females only, as applicable | X | | | | | | | | | | | | | | |
| Serum β-hCG[5] | X | X | | | | | | | | | | | | | X |
| IV Infusion Reaction Assessments[6] | | | | | X | -- | -- | -- | -- | -- | -- | -- | -- | -- | X |
| Pharmacodynamic/Pharmacokinetic Evaluations | | | | | | | | | | | | | | | |
| Blood for MK-3415A assay[7] | | | X | | X | X[7] | X | X | X | X | | | X | X | X |
| Other Study Procedures | | | | | | | | | | | | | | | |
| Study Drug Intravenous Infusion[8] | | | | X | -- | X | | | | | | | | | |
| Standardized Meals[9] | | | X | -- | -- | -- | -- | - | -- | -- | -- | -- | X | | |
| Domiciling in Clinical Research Unit[10] | | X | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | X | | |
| Evaluation of Adverse Experiences[11] | X | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | X |


| | | Study Day | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Pre-study[1] | -1 | 1 | | | | | | | | | | | 8 | Post-Study[1] |
| | | | | | Hours Post Initiation of Infusion | | | | | | | | | | |
| | | | Pre-dose | 0 | 0.5 | 1 | 2 | 4 | 6 | 8 | 12 | 16 | 24 | 168 | 504 |
| Study Procedure | | | | | | | | | | | | | | | |


1 Pre-study (screening) evaluations will be performed approximately 3 weeks from initial dose of study drug. Poststudy evaluations will be performed approximately 21 days after study drug administration.

2 A physical examination should be completed within 24 hours prior to study drug administration. Weight will be obtained with the subjects shoes off, jacket or coat removed, with a single calibrated scale. The Day -1 weight will be used by an unblinded pharmacist to calculate the amount of each monoclonal antibodies (based on the patient's weight in kilograms: 10 mg/kg of each monoclonal antibody) to add to a single bag of 0.9% sodium chloride to comprise a total infusion volume of 250 mL.

3 Triplicate ECGs (with at least a 1- to 2-minute interval between ECG measurements) will be conducted predose. All ECGs will be taken after the subject has remained in a semi-recumbent position for at least 10 minutes. All postdose ECGs are single measurements.

4 Subjects should be resting in a semi-recumbent position for at least 10 minutes prior to measurement of vital signs. Orthostatic vital signs will be obtained after the subject has been standing for at least 2 minutes. The predose (baseline) HR and BP (orthostatic and semi-recumbent) will be the average of duplicate measurements obtained at least 1-2 minutes apart within 30 minutes of dosing MK-3415A. Post-dose vital sign measurements will be single measurements.

5 Safety laboratory tests will be obtained following an 8-hour fast. The predose safety labs may be collected up to 24 hours prior to study drug administration. The Day -1 serum β-hCG must be confirmed negative prior to study drug administration.

6 Subjects will be monitored for infusion-specific events during the infusion and in the post-infusion monitoring period. See Section 3.2 for details.

7 Blood samples (3.5 mL) for determination of serum MK-3415A concentrations will be collected at the timepoints indicated. See Study Operations Manual for collection procedures. Sample collection at the 1-hour postdose timepoint should be collected immediately prior to the completion of the MK-3415A infusion. Blood samples for MK-3415A concentrations on Day 7 and Post-study can be collected within +/- 4 hours from specified time point.

8 MK-3415A or placebo will be administered as an IV infusion over 1 hour. Initiation of the infusion will be deemed t=0 and all postdose procedures will be based from this time. Study drug will be administered 1 hour after a standard breakfast on Day 1. See the Study Operations Manual for preparation and administration guidelines. Subjects will continue to rest semi-recumbent from dosing until 4 hours postdose except to stand for the measurement of orthostatic measurements or other study related procedure.

9 Standardized meals will be provided as follows: Day 1: breakfast 1 hr prior to dosing, lunch at ~4 hrs postdose, dinner at ~10 hrs postdose, and a snack will be offered at ~14 hrs postdose; other meals while domiciled in the CRU should be provided in a standardized fashion.

10 Subjects will remain in the CRU until 24 hours postdose and may be discharged at this time or at the discretion of the investigator.

11 Adverse experience monitoring to be initiated upon signing informed consent.

**Appendix 2.** Meso Scale Discovery Sector Imager 6000 (MSD) Electrochemiluminescent (ECL) Assay for the Measurement of Anti-MK-6072 Antibodies in Human Serum (Project RAOC) (Note: MK-6072: bezlotoxumab)

**Methods**

The assay for the detection of anti-MK-6072 in human serum was developed and validated at Merck, and transferred to (b) (4). Positive controls (PCs) and Negative controls (NCs), prepared in pooled human serum, as well as the samples were diluted and then loaded in duplicate onto an intermediary assay plate. Biotinylated-MK-6072 (capture antibody) and Ruthenylated-MK-6072 (detection antibody) with a final concentration of 0.10 μg/mL were added to all test wells of the intermediary assay plate. The capture antibody, ani-MK-6072, detection Ab complex was allowed to form during a 2-hour incubation period at room temperature. Anti-MK-6072 in the PCs and study samples was captured by the immobilized MK-6072 biotinylated antibody and detected by the ruthenylated MK-6072 antibody. Ruthenylated-MK-6072, in the presence of MSD read buffer, produced an electrochemiluminescent (ECL) signal, expressed in relative light units (RLU) that was proportional to the amount of anti-MK-6072 bound by the capture and detection reagents. A minimum sample volume of 25 μL is required.

The assay cut point was calculated for each plate. This floating cut point (FCP) in normal human serum was calculated, where the normalized cut point factor (NCF) determined during validation equals 5.95. During Tier 1 sample analysis, all study samples, including the pre-doses, were screened at the assay MRD of 1:2. Sample responses below the FCP are considered negative, while samples with responses at or above the FCP were considered potentially positive and are analyzed further to verify the specificity of the response.

Specificity of a potentially positive response was further evaluated during Tier 2 testing. A sample was considered positive for anti-MK-6072 antibodies if incubation with MK-6072 causes the sample response to fall above the specificity assay cut point or causes a reduction in signal greater than or equal to the specificity cut point in the spiked sample when compared to the unspiked sample. The fixed specificity cut point for normal serum was determined during validation to be 39.5. Positive samples were titered during Tier 3 testing. Samples that were determined to not be positive for anti-MK-6072 antibodies are not analyzed further and are considered negative.

**Results**

Analysis of human serum samples began on 12 January 2014 and was completed on 05 February 2014. The overall %CV for intra-assay precision samples ranged from 2.92% to 5.66%. The mean %CV for inter-assay precision samples for the high PC and low PC ranged from 17.5% to 18.1%. The overall limit of detection was 0.189 ng/mL. The addition of MK-3415 ranging from 1.00 to 10,000 ng/mL resulted in percent signal inhibition (%INH) from 0.00 to 15.6%, demonstrating that MK-3415 does not interfere with the assay up to 10,000 ng/mL. The addition of MK-6072 ranging from 1.00 to 10,000 ng/mL resulted in %INH from 1.04 to 99.6%. In a test using 22.2 ng/mL of anti-MK6072, the assay can still detect anti-MK6072 in the presence of 1000 ng/mL of MK-6072. The assay will use a floating cut point with a normalization factor of 5.95 for normal

individuals and 1.23 for disease state individuals. The specificity cut point was 39.5% for normal individuals and 34.6% for disease state individuals.

**Appendix 3**. Clinical Pharmacology Review for Meso Scale Discovery Sector Imager 6000 (MSD) Electrochemiluminescent (ECL) Method to Measure the Concentration of MK-6072 in Human Serum (Projects RAOB)
(Note: MK-6072: bezlotoxumab)

Streptavidin-coated plates are blocked with 1% BSA/1XPBST blocking buffer for up to 3 hours at ambient room temperature. Calibrators (CALs) and quality controls (QCs), and samples all in 10% pooled human serum are loaded onto an intermediary assay plate or into cluster tubes. Biotinylated anti-15B7 and Ruthenylated anti-15B7 are diluted to a final concentration of 0.75 µg/mL in matrix buffer and added to all test wells of the intermediary assay plate or into the cluster tubes. The capture Ab, MK-6072, detection Ab complex is allowed to form during a 1 to 3 hour incubation period at room temperature. After the incubation, the blocking buffer is decanted from the Streptavidin coated plate. The labeled anti-drug-MK-6072 complex is loaded onto the blocked plate and incubated up to 1.5 hours at ambient room temperature. MK-6072 in the CALs, QCs and study samples is captured by the biotinylated anti-15B7 antibody and detected by the ruthenylated anti-15B7 antibody. Unbound materials are removed from the plate by washing and 1X MSD read buffer is added to the wells. MSD read buffer produces an electrochemiluminescent (ECL) signal, expressed in relative light units (RLU) that is proportional to the amount of MK-6072 bound by the capture and detection reagents. The conversion of RLU for the samples and the QCs to concentrations is achieved through a LIMS system computer software mediated comparison to a standard curve on the same plate, which is regressed according to a 5-parameter logistic regression model with a weighting factor of $1/Y^2$: $y = d + ((a - d) / (1 + (x / c)^b)) (1/ratio^2)$. Results are reported in ng/mL concentration units.

**Results**
Analysis of human serum samples began on 20 December, 2013 and was completed on 21 February 2014. Each calibration curve was calculated using a five-parameter logistic ($1/response^2$ weighted) least-squares regression algorithm. Concentration data are reported to three significant figures. Back-calculated calibration standards are shown in **Table A3-1**. The average correlation coefficient for all obtained standard curve was 0.9985. Summary of Inter-assay Precision and Accuracy is shown in **Table A3-2**.

**Table A3-1**. Average Back-calculated Calibration Standards for Accepted Runs

| Run ID | CAL 1 (ng/mL) | CAL 2 (ng/mL) | CAL 3 (ng/mL) | CAL 4 (ng/mL) | CAL 5 (ng/mL) | CAL 6 (ng/mL) | CAL 7 (ng/mL) |
|---|---|---|---|---|---|---|---|
| N | (b) (4) | | | | | | |
| Theoretical Concentration | | | | | | | |
| Mean | | | | | | | |
| S.D. | | | | | | | |
| %C.V. | | | | | | | |
| % Difference from Theoretical | | | | | | | |

**Table A3-2**. Statistical Summary of Inter-assay Precision and Accuracy for Accepted Runs

| Run ID | QC 2 Dil 10 (ng/mL) | QC 3 Dil 10 (ng/mL) | QC 4 Dil 10 (ng/mL) |
|---|---|---|---|
| N | 90 | 90 | 89 |
| Theoretical Concentration | 300 | 800 | 5120 |
| Mean | 328 | 865 | 5540 |
| S.D. | 18.3 | 38.0 | 280 |
| %C.V. | 5.59 | 4.39 | 5.05 |
| % Difference from Theoretical | 9.29 | 8.14 | 8.22 |

**STUDY NO.: PROTOCOL 005-00 (P005)**

**Title: A Single Dose Study to Evaluate the Safety, Tolerability, and Pharmacokinetics of a 1-hour Intravenous Infusion of MK-3415A**

| | |
|---|---|
| **Date(s):** | 01 SEP 2010 – 05 JAN 2011 |
| **Investigator(s):** | Shwe Gyaw, M.D. Parexel International, Baltimore, MD, |
| **Study Center(s):** | Parexel International, Baltimore, MD |
| **Analytical Site(s):** | (b) (4) |

*Note: This study evaluated the PK of MK-3415A (a combination of monoclonal antibodies MK-3415 and MK-6072), but the current NDA submission intends to register MK-6072 only. Thus, the current Clinical Pharmacology review will focus on the study result of MK-6072.*

**OBJECTIVE(S):**
**Primary:** To evaluate the safety and tolerability of intravenous (IV) doses of MK-3415A administered over 1 hour by IV infusion.
**Secondary:** To obtain pharmacokinetic parameters (e.g., $AUC_{0\text{-last}}$, $C_{max}$, and $T_{max}$ in all subjects, plus $AUC_{0\text{-inf}}$ and $t_{½}$ in subjects in Cohort 1) following a 1 hour infusion of MK-3415A.

**METHODS**
**Study Design:** This is a double-blind, randomized, placebo-controlled, single dose study to evaluate the safety, tolerability, and pharmacokinetics of MK-3415A (monoclonal antibodies against *C. difficile* Toxin A and Toxin B). Thirty-five healthy male and female (of non-childbearing potential) subjects were allocated to this study; a subset of subjects (N=12) participated in a full assessment of pharmacokinetics until Day 85 (Cohort 1) and the remainder (N=23) participated until Day 22 (Cohort 2). All subjects received an intravenous (IV) infusion of MK-3415A (10 mg/kg of each monoclonal antibody to *C. difficile* Toxin A [MK-3415] and Toxin B [MK-6072]) or placebo as a 1 hour infusion on Day 1. Blood samples were collected predose and up to the poststudy visit from all subjects for pharmacokinetic (PK) analysis. Cohort 1(N=12) poststudy visit was conducted on Day 85. Cohort 2 (N=23) poststudy visit was conducted on Day 22. The duration of the study was approximately 15 weeks for Cohort 1 and approximately 6 weeks for Cohort 2 including prestudy and poststudy evaluations. The schedule of clinical observations and laboratory measurements is shown in **Appendix 1**.

Pharmacokinetics: Serum was analyzed pre- and postdose for MK-6072 concentrations. Study flow chart in **Appendix 1** shows the specific timepoints for sample collection. The serum pharmacokinetic parameters of MK-6072 were summarized by $AUC_{0\text{-last}}$, $C_{max}$, and $T_{max}$. For those subjects in whom a full pharmacokinetic profile was collected (Cohort 1), $AUC_{0\text{-inf}}$ and $t_{½}$ were also determined.

Safety: The safety and tolerability of MK-3415A were monitored by clinical assessment of adverse experiences and by repeated measurements of vital signs, physical examination, ECGs, and standard laboratory safety tests (hematology, serum chemistry and urinalysis). These

procedures were also performed at various unscheduled time points, if deemed clinically necessary by the investigator. The following infusion-specific events were prompted for during the infusion and throughout the study: infusion-site adverse experiences, pyrexia, chills, rash, arthralgia, joint swelling, obstructive airways disorder, bronchospasm, stridor, dysphonia, headache, fatigue, urticaria, hypotension, hypertension, nasal congestion, nausea, vomiting, flushing, angioedema, dyspnea, and dizziness/lightheadedness.

**Analytical Methods:**
Four-hundred nine (409) human serum samples were analyzed between 26 October 2010 and 26 May 2011. The bioanalytical assay using electrochemiluminescent (ECL) method to measure concentration of MK-6072 in human serum was performed by a CRO company (b) (4), (b) (4)). Please see **Appendix 2** for the detail of the ECL method. The analytical method was initially validated under project code "JBW2". During the course of the study, the method was updated and the validation was performed and also amended under project code "JBW4."

*Reviewer comments:*
*Overall, the bio-analytical method can specifically measure MK-6072 (Bezlotoxumab) concentration in human serum, and it is deemed appropriate after a Clinical Pharmacology review.*

**RESULTS**
**Study Population:** A subset of these (N=12) participated in a full assessment of pharmacokinetics through Day 85 (Cohort 1) and the remainder (N=23) participated until Day 22 (Cohort 2). All 35 subjects received an intravenous (IV) infusion of MK-3415A (10 mg/kg of each monoclonal antibody, MK-3415 and MK-6072, N=29) or placebo (N=6) over 1 hour on Day 1, in an infusion volume of 250 mL. Blood samples, for pharmacokinetic (PK) analysis, were collected predose and up to the poststudy visit for all subjects. The demographic information for the 35 subjects entered into the study is provided **Table 1**.

**Table 1**. Summary of Demographic Data

| Study Summary | Age (yr) | Height (cm) | Weight (kg) | BMI (kg/m$^2$) |
|---|---|---|---|---|
| Total N = 35: | | | | |
| Range: | 27 to 75 | 151.0 to 186.0 | 50.0 to 93.4 | 20.2 to 30.0 |
| Mean: | 48.8 | 172.5 | 77.5 | 26.1 |
| Male N = 28: | | | | |
| Male Range: | 27 to 70 | 165.0 to 186.0 | 62.2 to 93.4 | 20.2 to 30.0 |
| Male Mean: | 46.1 | 175.4 | 79.0 | 25.7 |
| Female N = 7: | | | | |
| Female Range: | 34 to 75 | 151.0 to 171.0 | 50.0 to 85.0 | 21.9 to 29.9 |
| Female Mean: | 59.4 | 160.9 | 71.5 | 27.5 |

**Pharmacokinetics:** Concentration-time profiles of MK-6072 from individual subject are depicted in **Figure 1**. Summary serum pharmacokinetic data for MK-6072 following a single IV dose of 10 mg/kg of each antibody infused over 1 hour are presented in **Table 2**. For comparison purposes, **Table 3** shows the pharmacokinetic parameters for Cohort 1 of the present study and

the 10 mg/kg MK-6072 arm of study P020 (CA-GCDX-05-01). Cohort 1 from the present study was used for the comparison because it contains data through Day 85. Study P020 was a rising single dose study of MK3415A in healthy subjects using a 2 hr IV infusion of 200 mL with data collection also through Day 85.

For MK-6072, observed antibody concentrations and associated PK parameters in P005 were approximately 10% lower than in Study P020. The observed variability in this study (P005) (about 15-25% CV) was similar to that observed in a prior study (P020).

**Figure 1.** Individual Serum Concentration Versus Time Profiles of MK-6072 Following Administration of a Single IV Dose of MK-3415A (10 mg/kg of each antibody) to Healthy Subjects (N=10 for Cohort 1, N=19 for Cohort 2) (Semi-log Scale)

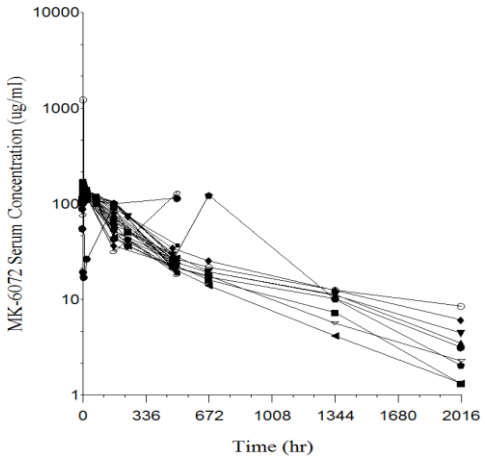



**Table 2**. Summary Statistics of Serum Pharmacokinetic Parameters for MK-6072 Following Administration of a Single IV Dose of MK-3415A (10 mg/kg of each antibody) as a 1-hr Infusion to Healthy Subjects (N=10 for Cohort 1, N=19 for Cohort 2)

| | Cohort | N | Mean (SD)† | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | | $AUC_{0\text{-last}}$ (µg·hr/mL) | $C_{max}$ (µg/mL) | $T_{max}$‡ (hr) | $AUC_{0-\infty}$ (µg·hr/mL) | Apparent Terminal $t_{1/2}$§ (hr) | V (L) | Cl (mL/min) |
| MK-6072 | 1 | 10 | 72200 (17700) | 404 (488) | 1.52 (0.98, 4.00) | 77600 (16800) | 424 (140) | 6.99 (1.83) | 0.179 (0.0391) |
| | 1* | 10 | 71900 (17600) | 248 (31) | 1.52 (0.98, 4.00) | 77400 (16700) | 424 (140) | 7.01 (1.83) | 0.179 (0.0385) |
| | 2 | 19 | 46100 (7270) | 237 (39) | 2.00 (0.98, 6.05) | NC | NC | NC | NC |
| | 1 & 2 | 29 | NA | 295 (290) | 2.00 (0.98, 6.05) | NC | NC | NC | NC |
| | 1 & 2* | 29 | NA | 241 (36) | 2.00 (0.98, 6.05) | NC | NC | NC | NC |

† Arithmetic mean and standard deviation based on the raw scale
‡ Median (Min, Max)
§ Harmonic Mean (Pseudo-SD)
* Cohort 1, Subject 1 had a serum MK-6072 concentration of 1790 µg/mL at 4 hr, which was ~8 fold higher than the 2 hr time point and ~7 fold higher than the mean Cmax of Cohort 1 calculated without the outlier. The mean PK parameters listed in the rows designated by * were calculated without this point for comparison
N = Number of subjects

**Table 3.** Comparison of Summary Statistics for Serum Pharmacokinetic Parameters for MK-3415 and MK-6072 Following IV Administration of 10 mg/kg MK-3415A from Study P005 Cohort 1 and P020

| | MK-3415 Mean (SD) | | MK-6072 Mean (SD) | |
|---|---|---|---|---|
| | P005 Cohort 1[1] | P020[2] MK-3415A 10 mg/kg | P005 Cohort 1[1] | P020[2] MK-3415A 10 mg/kg |
| N | 10 | 6 | 10 | 6 |
| AUC 0-inf (hr•µg/mL) | 115000 (21100) | 146000 (27100) | 77600 (16800) | 84400 (14100) |
| AUC 0-last (hr•µg/mL) | 97800 (19900) | 118000 (18700) | 72200 (17700) | 78300 (11400) |
| Cmax (µg/mL) | 261 (35) | 316 (19.9) | 404[3] (488) | 271 (52.7) |
| t1/2[4] (day) | 27.1 (5.20) | 32.5 (7.28) | 17.7 (5.80) | 21.3 (3.87) |

1: Cohort 1 from P005 was used for comparison because data were collected out to Day 85 which enables comparison with P020 which also collected data out to Day 85. Infusion time was 1 hr and infusion volume was 250 mL.
2: P020 is also known as CA-GCDX-05-10. Infusion time was 2 hr and infusion volume was 200 mL. PK parameters from P020 have been rounded to 3 significant figures.
3: The high Cmax value observed with MK-6072 is primarily due to one data point (Subject 1, 1790 µg/mL at 4 hrs) which was ~8 fold higher than the 2 hr time point. For comparison, analysis performed without this single point shows a mean and SD for Cmax of 248 (31) for cohort 1.
4: Half-life was originally reported in hours but is converted to days for comparison. For P005 t ½ was reported as Harmonic Mean (Pseudo-SD). For P020 t ½ was reported as arithmetic mean and standard deviation.

*Reviewer Comment:*

1) *Interestingly, the half-life of MK-3415 is longer than MK-6072, although both of them are similar IgGs.*
2) *The PK parameters of MK-6072 appear adequately determined in this study.*
3) *The PK result from Study P020 is presented for the purpose of reference, due to nonspecific bioanalytical assay used in Study P020.*

**Safety:**

All 35 enrolled subjects were included in the assessment of safety and tolerability. Eleven (31%) subjects reported a total of 15AEs after treatment; the majority of AEs were considered by the investigator to be mild in intensity. All except one AE (headache) which occurred in the immediate post-infusion monitoring period were considered not related to study drug. Of the 15 non-serious clinical adverse events, the most common were excoriation (2 subjects with 1 event each) and back pain (2 subjects with 1 event each). No AE required medical intervention apart from the administration of concomitant medication. There were no serious clinical, laboratory, or other AEs and no subjects died during the study.

**SPONSOR'S CONCLUSIONS:**
MK-3415A [MK-3415 and MK-6072] is a combination of monoclonal antibodies against toxins A and B from *Clostridium difficile*. MK-3415A is being developed for the prevention of recurrence of *C. difficile* infection. Prior to this study, MK-6072 had been evaluated in clinical trials after infusions of no less than two hours and in volumes of 200 mL. This study is the first study to assess the PK and tolerability profile of MK-3415A administered over one hour and with a 250 mL infusion.

As noted, MK-3415A was generally well tolerated. In the acute, peri-infusional phase, there were no discontinuations or infusion slowings due to adverse events. In the chronic, post-infusional phase, there were no serious adverse events. The adverse events reported were generally similar to those in the placebo group.

Overall, the results of this study suggest that the pharmacokinetics of MK-6072 after 1 hour and 2 hour infusions (Study P020) is broadly comparable, within ~25%. For MK-6072, the observed variability in Study P005 (~15-25% CV) was similar to that observed in Study P020. Differences in the observed pharmacokinetics of these antibodies are likely to be due to study-to-study variation and differences between the assays used to assess antibody concentrations between the present study and Study P020. Differences in length of infusion or volume of administration are unlikely to be clinically meaningful.

The results of this study support the clinical use of MK-3415A at a dose of 10 mg/kg of MK-3415 and MK-6072 administered over 1 hour in a 250 mL infusion volume.

**REVIEWER ASSESSMENT:**
Following a single dose of 10 mg/kg of MK-3415A (a combination of MK-3415 and MK-6072) administered over 1 hour, the PK parameters of MK-6072 were determined adequately. The safety profile appears acceptable.

## Appendix 1.

## Table A1-1. Study Flow Chart for Cohort 1

| | | Study Day | | | | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Pre-Study[1] | -1 | 1 | | | | | | | | | | 2 | 4 | 8 | 11 | 21 | 29 | 57 | Post-study[1] |
| | | | | Hour Post Initiation of Infusion | | | | | | | | | | | | | | | | |
| | | | Pre-dose | 0 | 0.5 | 1 | 2 | 4 | 6 | 8 | 12 | 16 | 24 | 72 | 168 | 240 | 480 | 672 | 1344 | 2016 |
| Study Procedure | | | | | | | | | | | | | | | | | | | | |
| Study informed consent | X | | | | | | | | | | | | | | | | | | | |
| Medical History/Assessment of Inclusion/Exclusion Criteria | X | | | | | | | | | | | | | | | | | | | |
| HIV/Hepatitis screen (per site SOP) | X | | | | | | | | | | | | | | | | | | | |
| Drug and alcohol screen (per site SOP) | X | X | | | | | | | | | | | | | | | | | | |
| Genetic Informed Consent | | X | | | | | | | | | | | | | | | | | | |
| Genetic Sample Collection | | | X | | | | | | | | | | | | | | | | | |
| Safety evaluations | | | | | | | | | | | | | | | | | | | | |
| Physical examination (PE)[2] | X | | X | | | | | | | | | | | | | | X | | | X |
| Weight[2] | X | X | | | | | | | | | | | | | | | X | | | X |
| Height | X | | | | | | | | | | | | | | | | | | | |
| 12-lead electrocardiogram (ECG)[3] | X | | X | | | X | X | | | X | X | | X | | | | X | | | X |
| Semi recumbent vital signs (HR, BP, RR, oral T)[4] | X | | X | | X | X | X | X | | X | X | X | X | | X | X | X | X | X | X |
| Orthostatic vital signs (HR, BP)[4] | | | X | | | X | | | | | | | | | | | | | | |
| Laboratory safety tests (serum chemistry, CBC, urinalysis)[5] | X | | X | | | | | | | | | | X | | | | X | | | X |
| FSH, females only, as applicable | X | | | | | | | | | | | | | | | | | | | |
| Serum β-hCG[5] | X | X | | | | | | | | | | | | | | | | | | X |
| IV Infusion Reaction Assessments[6] | | | | | X | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | X |
| Pharmacodynamic/Pharmacokinetic Evaluations | | | | | | | | | | | | | | | | | | | | |
| Blood for MK-3415A assay[7] | | | X | | X | X[7] | X | X | X | X | | | X | X | X | X | X | X | X | X |
| Other Study Procedures | | | | | | | | | | | | | | | | | | | | |
| Study Drug Intravenous Infusion[8] | | | | X | -- | X | | | | | | | | | | | | | | |
| Standardized Meals[9] | | | X | -- | -- | -- | -- | - | -- | -- | -- | -- | X | | | | | | | |
| Domiciling in Clinical Research Unit[10] | | X | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | X | | | | | | | |
| Evaluation of Adverse Experiences[11] | X | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | X |
| | | Study Day | | | | | | | | | | | | | | | | | | |

| | Pre-Study[1] | -1 | 1 | | | | | | | | | | 2 | 4 | 8 | 11 | 21 | 29 | 57 | Post-study[1] |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | Hour Post Initiation of Infusion | | | | | | | | | | | | | | | | |
| | | | Pre-dose | 0 | 0.5 | 1 | 2 | 4 | 6 | 8 | 12 | 16 | 24 | 72 | 168 | 240 | 480 | 672 | 1344 | 2016 |
| Study Procedure | | | | | | | | | | | | | | | | | | | | |

1 Pre-study (screening) evaluations will be performed approximately 3 weeks from initial dose of study drug. Poststudy evaluations will be performed approximately 84 days after study drug administration.

2 A physical examination should be completed within 24 hours prior to study drug administration. Weight will be obtained with the subjects shoes off, jacket or coat removed, with a single calibrated scale. The Day -1 weight will be used by an unblinded pharmacist to calculate the amount of each monoclonal antibodies (based on the patient's weight in kilograms: 10 mg/kg of each monoclonal antibody) to add to a single bag of 0.9% sodium chloride to comprise a total infusion volume of 250 mL.

3 Triplicate ECGs (with at least a 1- to 2-minute interval between ECG measurements) will be conducted predose. All ECGs will be taken after the subject has remained in a semi-recumbent position for at least 10 minutes. All postdose ECGs are single measurements.

4 Subjects should be resting in a semi-recumbent position for at least 10 minutes prior to measurement of vital signs. Orthostatic vital signs will be obtained after the subject has been standing for at least 2 minutes. The predose (baseline) HR and BP (orthostatic and semi-recumbent) will be the average of duplicate measurements obtained at least 1-2 minutes apart within 30 minutes of dosing MK-3415A. Post-dose vital sign measurements will be single measurements.

5 Safety laboratory tests will be obtained following an 8-hour fast. The predose safety labs may be collected up to 24 hours prior to study drug administration. The Day -1 serum β-hCG must be confirmed negative prior to study drug administration.

6 Subjects will be monitored for infusion-specific events during the infusion and in the post-infusion monitoring period. See Section 3.2 for details.

7 Blood samples (3.5 mL) for determination of serum MK-3415A concentrations will be collected at the timepoints indicated. See Study Operations Manual for collection procedures. Sample collection at the 1-hour postdose timepoint should be collected immediately prior to the completion of the M-3415A infusion. Blood samples for MK-3415A concentrations on Day 8 through Poststudy can be collected within +/- 4 hours from specified time point.

8 MK-3415A or placebo will be administered as an IV infusion over 1 hour. Initiation of the infusion will be deemed t=0 and all postdose procedures will be based from this time. Study drug will be administered 1 hour after a standard breakfast on Day 1. See the Study Operations Manual for preparation and administration guidelines. Subjects will continue to rest semi-recumbent from dosing until 4 hours postdose except to stand for the measurement of orthostatic measurements or other study related procedure.

9 Standardized meals will be provided as follows: Day 1: breakfast 1 hr prior to dosing, lunch at ~4 hrs postdose, dinner at ~10 hrs postdose, and a snack will be offered at ~14 hrs postdose; other meals while domiciled in the CRU should be provided in a standardized fashion.

10 Subjects will remain in the CRU until 24 hours postdose and may be discharged at this time or at the discretion of the investigator.

11 Adverse experience monitoring to be initiated upon signing informed consent.

## Table A1-2 Study Flow Chart Cohort 2

| | | Study Day | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Pre-study[1] | -1 | 1 | | | | | | | | | | | 8 | Post-Study[1] |
| | | | | | Hours Post Initiation of Infusion | | | | | | | | | | |
| | | | Pre-dose | 0 | 0.5 | 1 | 2 | 4 | 6 | 8 | 12 | 16 | 24 | 168 | 504 |
| Study Procedure | | | | | | | | | | | | | | | |
| Study informed consent | X | | | | | | | | | | | | | | |
| Medical History/Assessment of Inclusion/Exclusion Criteria | X | | | | | | | | | | | | | | |
| HIV/Hepatitis screen(per site SOP) | X | | | | | | | | | | | | | | |
| Drug and alcohol screen (per site SOP) | X | X | | | | | | | | | | | | | |
| Genetic Informed Consent | X | | | | | | | | | | | | | | |
| Genetic Sample Collection | | | X | | | | | | | | | | | | |
| Safety evaluations | | | | | | | | | | | | | | | |
| Physical examination (PE)[2] | X | | X | | | | | | | | | | | | X |
| Weight[2] | X | X | | | | | | | | | | | | | X |
| Height | X | | | | | | | | | | | | | | |
| 12-lead electrocardiogram (ECG)[3] | X | | X | | | X | X | | | X | X | | X | | X |
| Semi recumbent vital signs (HR, BP, RR, oral T)[4] | X | | X | | X | X | X | X | | X | X | X | X | | X |
| Orthostatic vital signs (HR, BP)[4] | | | X | | | X | | | | | | | | | |
| Laboratory safety tests (serum chemistry, CBC, urinalysis)[5] | X | | X | | | | | | | | | | X | | X |
| FSH, females only, as applicable | X | | | | | | | | | | | | | | |
| Serum β-hCG[5] | X | X | | | | | | | | | | | | | X |
| IV Infusion Reaction Assessments[6] | | | | | X | -- | -- | -- | -- | -- | -- | -- | -- | -- | X |
| Pharmacodynamic/Pharmacokinetic Evaluations | | | | | | | | | | | | | | | |
| Blood for MK-3415A assay[7] | | | X | | X | X[7] | X | X | X | X | | | X | X | X |
| Other Study Procedures | | | | | | | | | | | | | | | |
| Study Drug Intravenous Infusion[8] | | | | X | -- | X | | | | | | | | | |
| Standardized Meals[9] | | | X | -- | -- | -- | -- | - | -- | -- | -- | -- | X | | |
| Domiciling in Clinical Research Unit[10] | | X | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | X | | |
| Evaluation of Adverse Experiences[11] | X | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | -- | X |


| | | Study Day | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Pre-study[1] | -1 | 1 | | | | | | | | | | | 8 | Post-Study[1] |
| | | | | | Hours Post Initiation of Infusion | | | | | | | | | | |
| | | | Pre-dose | 0 | 0.5 | 1 | 2 | 4 | 6 | 8 | 12 | 16 | 24 | 168 | 504 |
| Study Procedure | | | | | | | | | | | | | | | |


1 Pre-study (screening) evaluations will be performed approximately 3 weeks from initial dose of study drug. Poststudy evaluations will be performed approximately 21 days after study drug administration.

2 A physical examination should be completed within 24 hours prior to study drug administration. Weight will be obtained with the subjects shoes off, jacket or coat removed, with a single calibrated scale. The Day -1 weight will be used by an unblinded pharmacist to calculate the amount of each monoclonal antibodies (based on the patient's weight in kilograms: 10 mg/kg of each monoclonal antibody) to add to a single bag of 0.9% sodium chloride to comprise a total infusion volume of 250 mL.

3 Triplicate ECGs (with at least a 1- to 2-minute interval between ECG measurements) will be conducted predose. All ECGs will be taken after the subject has remained in a semi-recumbent position for at least 10 minutes. All postdose ECGs are single measurements.

4 Subjects should be resting in a semi-recumbent position for at least 10 minutes prior to measurement of vital signs. Orthostatic vital signs will be obtained after the subject has been standing for at least 2 minutes. The predose (baseline) HR and BP (orthostatic and semi-recumbent) will be the average of duplicate measurements obtained at least 1-2 minutes apart within 30 minutes of dosing MK-3415A. Post-dose vital sign measurements will be single measurements.

5 Safety laboratory tests will be obtained following an 8-hour fast. The predose safety labs may be collected up to 24 hours prior to study drug administration. The Day -1 serum β-hCG must be confirmed negative prior to study drug administration.

6 Subjects will be monitored for infusion-specific events during the infusion and in the post-infusion monitoring period. See Section 3.2 for details.

7 Blood samples (3.5 mL) for determination of serum MK-3415A concentrations will be collected at the timepoints indicated. See Study Operations Manual for collection procedures. Sample collection at the 1-hour postdose timepoint should be collected immediately prior to the completion of the MK-3415A infusion. Blood samples for MK-3415A concentrations on Day 7 and Post-study can be collected within +/- 4 hours from specified time point.

8 MK-3415A or placebo will be administered as an IV infusion over 1 hour. Initiation of the infusion will be deemed t=0 and all postdose procedures will be based from this time. Study drug will be administered 1 hour after a standard breakfast on Day 1. See the Study Operations Manual for preparation and administration guidelines. Subjects will continue to rest semi-recumbent from dosing until 4 hours postdose except to stand for the measurement of orthostatic measurements or other study related procedure.

9 Standardized meals will be provided as follows: Day 1: breakfast 1 hr prior to dosing, lunch at ~4 hrs postdose, dinner at ~10 hrs postdose, and a snack will be offered at ~14 hrs postdose; other meals while domiciled in the CRU should be provided in a standardized fashion.

10 Subjects will remain in the CRU until 24 hours postdose and may be discharged at this time or at the discretion of the investigator.

11 Adverse experience monitoring to be initiated upon signing informed consent.

**Appendix 2**. Clinical Pharmacology Review for Meso Scale Discovery Sector Imager 6000 (MSD)Electrochemiluminescent (ECL) Assay for the Measurement of MK-6072 in Human Serum (Projects WJW, JBW2, and JBW4)

**Methods**

Streptavidin-coated plates were blocked with 1% BSA/1XPBST blocking buffer for up to 3 hours at ambient room temperature. Calibrators (CALs) and quality controls (QCs), and samples all in 10% pooled human serum are loaded onto an intermediary assay plate or into cluster tubes. Biotinylated anti-15B7 and Ruthenylated anti-15B7 were diluted to a final concentration of 0.75 µg/mL in matrix buffer and added to all test wells of the intermediary assay plate or into the cluster tubes. The capture Ab, MK-6072, detection Ab complex is allowed to form during a 1 to 3 hour incubation period at room temperature. After the incubation, the blocking buffer is decanted from the Streptavidin coated plate. The labeled anti-drug-MK-6072 complex is loaded onto the blocked plate and incubated up to 1.5 hours at ambient room temperature. MK-6072 in the CALs, QCs and study samples is captured by the biotinylated anti-15B7 antibody and detected by the ruthenylated anti-15B7 antibody. Unbound materials were removed from the plate by washing and 1X MSD read buffer is added to the wells. MSD read buffer produces an ECL signal, expressed in relative light units (RLU) that is proportional to the amount of MK-6072 bound by the capture and detection reagents.

The QCs were prepared by diluting the MK-6072 reference standard in 100% pooled human serum. The QC concentration levels included 100 (QC1, LLOQ), 300 (QC2, low QC), 800 (QC3, mid QC), 5120 (QC4, high QC), 6400 (QC5, ULQC), 44000 (QC6, Dilution QC), 600000 (QC7, Dilution QC), 800000 ng/mL (QC8, Dilution QC).

**Results**

Analysis of human serum samples began on 10 December 2010 and was completed on 02 June 2011. The conversion of RLU for the samples and the QCs to concentrations is achieved through a LIMS system computer software mediated comparison to a standard curve on the same plate, which is regressed according to a 5-parameter logistic (5-PL) regression model with a weighting factor of $1/Y^2$: $y = d + ((a - d) / (1 + (x / c)b)) (1/ratio^2)$. The average correlation coefficient for all obtained standard curves was 0.9988. Back-calculated calibration standards are shown in **Table A2-1**. The inter-assay precision and accuracy for the standard calibrators ranged from 0.934 to 1.85% and -2.42 to 3.06 %, respectively

**Table A2-1**. Average Back-calculated Calibration Standards (Concentration in ng/mL)

| | (b) (4) |
|---|---|
| N | |
| Theoretical Concentration | |
| Mean | |
| S.D. | |
| %C.V. | |
| % Difference From Theoretical | |

The in-study inter-assay precision and accuracy were evaluated by replicate analyses of human serum QC pools prepared at three concentrations spanning the calibration range (**Table A2-2**). Precision ranged from 6.91 to 7.85%, measured as the percent coefficient of variation (%CV) of the set of values for each pool. Accuracy ranged from -6.44 to -5.65% and was expressed as the percent difference of the mean value for each pool from the theoretical concentration.

**Table A2-2**. Inter-assay Precision and Accuracy

| | | | |
|---|---|---|---|
| N | 116 | 116 | 116 |
| Concentration | 300 | 800 | 5120 |
| Mean | 281 | 748 | 4830 |
| S.D. | 22.1 | 52.9 | 334 |
| %C.V. | 7.85 | 7.06 | 6.91 |
| from Theoretical | -6.17 | -6.44 | -5.65 |

**Validation Results**

The Inter-assay precision and accuracy was obtained from QC1, QC2, QC3, QC4, and QC5 on each test plate. The assay was accurate from 100ng/mL to 6400 ng/mL with per plate intra-assay accuracy ranging from -21.6% to 8.05% and overall inter-assay accuracy from -10.4% to 0.453%. The assay was precise from 100 ng/mL to 6400 ng/mL. The per plate intra-assay precision ranged from 0.425% to 29.2% and overall inter-assay precision from 4.35% to 13.1%. The intra-assay and inter-assay precisions were acceptable based on the predefined acceptance criteria. The dilutional linearity test using QC6 (44000 ng/mL) showed that the global percent difference from theoretical concentration (% Bias) was ≤ 11.9% for all valid dilutions (up to 2560 fold dilution). The range of calibration was 10.0 ng/mL to 640.0 ng/mL. The range of quantitation was 100 ng/mL to 6400 ng/mL. The required sample volume for analysis was 10.0 µL.

Accuracy and precision of a TECAN EVO workstation to perform sample dilutions beyond the calibration curve range as well as the minimum required dilution (MRD) for the ECL immunoassay on the Sector Imager 6000 (SI6000) from MSD were evaluated utilizing quality control (QCs) at four concentrations: 300ng/mL (LQC), 800 ng/mL (MQC), 5120 ng/mL (HQC), and 600000 ng/mL (DQC). The percent difference from theoretical (%Bias, PDT) of the mean of each of the DQC dilution factors demonstrated accuracy and precision were acceptable (within 20% of the theoretical concentration). The partially automated assay proved accurate across all dilution factors with PDTs ranging from -16.4% to 4.37% with all runs reported. The intra-assay accuracy ranged from -12.0% to 10.4%. The inter-assay accuracy ranged from -5.05% to 0.924%. The demonstrated intra-assay precision ranged from 1.35% to 9.46% across the various dilution factors. The inter-assay precision of the plate acceptance QCs ranged from 5.99% to 8.09%.

The results of storage stability at 2° to 8° C are shown in **TableA2-3**.The LQC, MQC, and HQC (RF2-4, respectively) were acceptable with accuracy of the mean value of -5.57% to -0.197% and precision of 4.92% to 7.39%.The 1:4000 DQC (RF7) was acceptable with accuracy of 6.09% and precision of 2.82%.

**Table A2-3** Twenty-four hour Storage Stability (2-8 ºC) for MK-6072 in Human Serum

| Date/Run ID/Analyst | RF 2<br>Dil 10 | RF 3<br>Dil 10 | RF 4<br>Dil 10 | RF 7<br>Dil<br>40000 |
|---|---|---|---|---|
| 10DEC2010/2JBW4/ (b) (4) | 312<br>303<br>283 | 819<br>715<br>733 | 5210<br>4750<br>4800 | 654000<br>638000<br>618000 |
| N | 3 | 3 | 3 | 3 |
| Theoretical Concentration | 300 | 800 | 5120 | 600000 |
| Mean | 299 | 755 | 4920 | 637000 |
| S.D. | 14.7 | 55.8 | 254 | 18000 |
| %C.V. | 4.92 | 7.39 | 5.17 | 2.82 |
| % Difference from Theoretical | -0.197 | -5.57 | -3.95 | 6.16 |

RF: Refrigerated (2-8ºC) Stability Sample

In summary, the assay showed dilutional linearity between dilutions 160 to 2560 from a QC at 44000 ng/mL after the MRD in matrix buffer. Samples were stable for up to 24 hours at 2° to 8°C up to a dilution of 40000. The TECAN EVO workstation was proven as acceptable for use to perform sample dilutions beyond the curve range and the MRD. All dilution schemes tested were validated for use.

**STUDY NO.: PROTOCOL 006-00 (P006)**

**Title: A Single Dose Study to Evaluate the Safety, Tolerability, and Pharmacokinetics of MK-3415A in Healthy Japanese Male Subjects**

**Date(s):** 07 SEP 2011– 19 APR 2012
**Investigator(s):** Hakop Gevorkyan, MD, MBA, PAREXEL International Early Phase Clinical Unit, Glendale, CA
**Study Center(s):** Parexel International, Baltimore, MD
**Analytical Site(s):** (b) (4)

*Note: This study evaluated the PK of MK-3415A (a combination of monoclonal antibodies MK-3415 and MK-6072), but the current NDA submission intends to register MK-6072 only. Thus, the current Clinical Pharmacology review will focus on the study result of MK-6072.*

**OBJECTIVE(S):**
**Primary:** To evaluate the safety and tolerability of single intravenous (IV) doses of MK-3415A in healthy adult Japanese male subjects.

**Secondary:** To obtain serum PK (e.g., $AUC_{0\text{-last}}$, $AUC_{0\text{-}\infty}$, $T_{max}$, $C_{max}$ and apparent terminal t½) of MK-6072 following single IV doses of MK-3415A in healthy adult Japanese male subjects.

**Exploratory:** To make a preliminary assessment of the immunogenicity of single IV doses of MK-3415A in healthy adult Japanese male subjects.

**METHODS**

**Study Design:** This was a double-blind, randomized, placebo-controlled, single dose study to evaluate the safety, tolerability, and pharmacokinetics of two doses of MK-3415A (monoclonal antibodies to *C. difficile* Toxin A and Toxin B) in healthy Japanese male volunteers:
In Panel A, subjects received a single dose of 10 mg/kg MK-3415A (10 mg/kg MK-3415 + 10 mg/kg MK-6072) or placebo. In Panel B, subjects received a single dose of 20 mg/kg MK-3415A (20 mg/kg MK-3415 + 20 mg/kg MK-6072) or placebo. The amount of each monoclonal antibody was calculated and added to a single bag of 0.9% sodium chloride to comprise a final total infusion volume of 250 mL. Study infusions were administered at a constant rate in no less than 1 hour. The schedule of clinical observations and laboratory measurements is shown in **Table 1**.

**Table 1**. Study Flow Chart

| Study Procedures | Pre-study [1] | Study Day: -1 | Pre-dose | 1: 0 | 0.5 | 1 | 2 | 4 | 6 | 8 | 12 | 16 | 2: 24 | 4: 72 | 8: 168 | 11: 240 | 21: 480 | 29: 672 | 57: 1344 | 85 [14]: 2016 | 168 [1] (Post-study): 4008 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | Hour Post Initiation of Infusion | | | | | | | | | | | | | | | | | |
| Study informed consent | X | | | | | | | | | | | | | | | | | | | | |
| MH/assessment of incl./excl. criteria | X | | | | | | | | | | | | | | | | | | | | |
| Informed consent for FBR | X | | | | | | | | | | | | | | | | | | | | |
| Blood for FBR [13] | | | X | | | | | | | | | | | | | | | | | | |
| HIV/Hepatitis screen (per site SOP) | X | | | | | | | | | | | | | | | | | | | | |
| Drug and alcohol screen (per site SOP) | X | X | | | | | | | | | | | | | | | | | | | |
| **Safety evaluation** | | | | | | | | | | | | | | | | | | | | | |
| Physical examination [2] | X | X | | | | | | | | | | | | | X | | X | | | X | X |
| Weight [2] | X | X | | | | | | | | | | | | | | | X | | | X | X |
| Height | X | | | | | | | | | | | | | | | | | | | | |
| 12-Lead ECG [3] | X | | X | | | X | X | | | X | X | | X | | | | X | | | X | X |
| Vital signs (HR, BP, RR, BT) [4] | X | | X | | X | X | X | X | | X | X | X | X | X | X | X | X | X | X | X | X |
| Orthostatic vital signs (HR, BP) [4] | | | X | | | X | | | | | | | | | | | | | | | |
| Laboratory safety tests (serum chemistry, CBC, urinalysis) [5] | X | | X | | | | | | | | | | X | | | | X | | | X | X |
| IV infusion reaction assessment [6] | | | | | X---------- | | | | | | | | | | | | | | X | | |
| **Pharmacodynamic/pharmacokinetic evaluations** | | | | | | | | | | | | | | | | | | | | | |
| Blood for MK-3415A assay [7] | | | X | | X | X[7] | X | X | X | X | | | X | X | X | X | X | X | X | X | X |
| Blood sample for ADA level [12] | | | X | | | | | | | | | | | | | | X | | | X | X |
| **Other study procedures** | | | | | | | | | | | | | | | | | | | | | |
| Study drug IV infusion [8] | | | | X---------- | | X | | | | | | | | | | | | | | | |
| Standardized meals [9] | | | X---------- | | | | | | | | | | X | | | | | | | | |
| Domiciling in CRU | | X---------- | | | | | | | | | | | X | | | | | | | | |
| Evaluation of AEs/concomitant medication [11] | | | X---------- | | | | | | | | | | | | | | | | | X | X |

ADA = anti-drug antibody; BP = blood pressure; BT = body temperature; CRU = Clinical Research Unit; ECG = electrocardiogram; FBR = future biomedical research; HR = heart rate; MH = medical history; IV = intravenous; RR = respiratory rate; SOP = standard operating procedure

1 Pre-study (screening) evaluations were performed approximately 3 weeks from initial dose of study drug. Day 168 (poststudy) evaluations were performed approximately 168 days after study drug administration.

2 A physical examination was completed within 24 hours prior to study drug administration. Weight was obtained with the subjects shoes off, jacket or coat removed, with a single calibrated scale. The Day -1 weight was used by an unblinded pharmacist to calculate the amount of each monoclonal antibodies (based on the subject's weight in kilograms: 10 and 20 mg/kg of each monoclonal antibody) to add to a single bag of 0.9% sodium chloride to comprise a total infusion volume of 250 mL.

3 Triplicate ECGs (with at least a 1- to 2-minute interval between ECG measurements) were conducted predose. All ECGs were taken after the subject had remained in a semi-recumbent position for at least 10 minutes. All postdose ECGs were single measurements.

4 Subjects were resting in a semi-recumbent position for at least 10 minutes prior to measurement of vital signs. Orthostatic vital signs were obtained after the subject had been standing for at least 2 minutes. The predose (baseline) HR and BP (orthostatic and semi-recumbent) was the average of duplicate measurements obtained at least 1-2 minutes apart within 60 minutes of dosing MK-3415A. Post-dose vital sign measurements was single measurements.

5 Safety laboratory tests were obtained following an 8-hour fast. The predose safety laboratory assessments were collected up to 24 hours prior to study drug administration.

6 Subjects were monitored for infusion-specific events during the infusion and in the post-infusion monitoring period.

7 Blood samples (3.5 mL) for determination of serum MK-3415A concentrations were collected at the time points indicated (infusion start t=0). Sample collection at the 1-hour postdose time point were collected immediately after the completion of the M-3415A infusion. The exact draw time was recorded in the case report form. Blood samples for MK-3415A concentrations on Day 8 through Day 168 (poststudy) were collected within +/- 4 hours from specified time point.

8 MK-3415A or placebo was administered as an IV infusion over 1 hour. Initiation of the infusion was deemed t=0 and all postdose procedures based from this time. Study drug was administered 1 hour after a standard breakfast on Day 1. Subjects continued to rest semi-recumbent from dosing until 4 hours postdose except to stand for the measurement of orthostatic measurements or other study related procedure.

9 Standardized meals on Day 1 were provided 1 hr prior to dosing (breakfast), ~4 hrs postdose (lunch), and ~10 hrs postdose (dinner). A 30-minute allowance was deemed appropriate.

10 Subjects were discharged after the completion of all procedures on Day 2 (at the discretion of the investigator).

11 AEs were monitored until the final evaluation point of immunogenicity [6 months (168 days) after administration].

12 Blood samples (10 mL) for determination of serum MK-3415A concentrations were collected at the time points indicated.. Samples testing positive for ADA was then to be tested for neutralizing antibody.

13 Informed consent for future biomedical research samples was obtained before the DNA sample. DNA sample for analysis was obtained predose, on Day 1 (or with the next scheduled blood draw), as the last sample drawn, on randomized subjects only, or at a later date as soon as the informed consent was obtained. If sample was collected on a Friday, instructions for refrigeration in manual was to be followed. It was preferred to ship whole blood sample within 24 hours.

14 Day 85 was a critical visit (time point); all samples must be collected on this day (no time deviation was allowed).

Pharmacokinetics: Blood samples for PK analysis were obtained up to Day 168 after MK-3415A dosing. Pharmacokinetic parameters were determined for MK-6072, including $AUC_{0\text{-last}}$, $AUC_{0\text{-}\infty}$, $T_{max}$, $C_{max}$, CL, V, and apparent terminal t½ following single IV doses of MK-3415A. The geometric means (with 95% confidence interval [CI]) of MK-6072 for $AUC_{0\text{-}\infty}$, $AUC_{0\text{-last}}$ and $C_{max}$ at each treatment were assessed in order to explore the PK/dose relationship.

Immunogenicity: Blood samples for immunogenicity analysis were obtained up to Day 168 after MK-3415A dosing. Anti-drug antibody (ADA) was tested for MK-6072. If a sample at a given time point provided a confirmed positive ADA result(s), the sample was also tested for neutralizing antibody using a cell-based Neutralizing Antibody Assay.

Pharmacogenetics: Exploratory pharmacogenetic (PG) studies may have been performed if significant variability in pharmacokinetic concentrations was observed or AEs were identified. Genomic markers of disease may also have been investigated. Any significant PG relationships to outcome required validation in future clinical trials. No further exploration of pharmacogenomics samples were deemed necessary and were not completed.

Safety: Safety and tolerability were monitored by AE reporting, vital signs (including orthostatic assessment), ECGs, and safety laboratory assessments.

**Dosage/Formulation.:**
MK-3415A (combination MK-3415 + MK-6072), sterile solution for IV infusion. Drug supplies were provided as glass vials containing 25 mg/mL MK-3415 and 25 mg/mL MK-6072, respectively.
– Treatment A (Panel A): 10 mg/kg MK-3415A (i.e. 10 mg/kg MK-3415 + 10 mg/kg MK-6072) or placebo
– Treatment B (Panel B): 20 mg/kg MK-3415A (i.e. 20 mg/kg MK-3415 + 20 mg/kg MK-6072) or Placebo

| Drug | Potency | Dosage Form | Control No. / Formulation No. | Assay Potency (ELIZA / in vitro neutralization) | Site of Manufacture |
|---|---|---|---|---|---|
| MK-3415 | 25 mg / 40 mL | IV vial | WL00044409 / WL00036089 | (b) (4) | Merck: West Point, PA |
| MK-6072 | 25 mg / 40 mL | IV vial | WL00044410 / WL00036090 | | Merck: West Point, PA |
| ELIZA = enzyme-linked immunosorbent assay; IV = intravenous<br>Placebo (saline infusion bags) (b) (4) was sourced locally (b) (4). Batch/Lot number: C846030. Expiry date: Feb-2013. | | | | | |

**Analytical Methods:**
Whole blood samples were collected into Serum Separator Tubes (SST) for the analysis of MK-3415A serum concentrations at the protocol-specified time points. Serum samples were analyzed for MK-6072 and anti-MK-6072-antibodies. The analytical methods for the quantitation of MK-6072 and anti-MK-6072-antibodies were based on an electrochemiluminescent (ECL) immunoassay method using a Meso Scale Discovery Sector Imager 6000 (MSD). This method was validated by Merck Research Laboratories and (b) (4). under Project Codes "JBW2,", "JBW4", and "JBW5".

The lower limit of quantitation (LLOQ) was 100 ng/mL for MK-6072 and 0.189 ng/mL for anti-MK-6072, with a linear calibration range from 10 to 640 ng/mL for MK-6072. Please see **Appendix 1** for details.

*Reviewer comments:*
*Overall, the bio-analytical method can specifically assess MK-6072 (Bezlotoxumab) concentration in human serum, and it is deemed appropriate after a Clinical Pharmacology review. Details of the analytical method for MK-6072 and anti-MK-6072 antibody quantitation and their respective validations are presented in the bioanalytical and method validation reports.*

**RESULTS**
**Study Population:** Nineteen (19) healthy Japanese male subjects were enrolled in this study and 16 (84.2%) completed the study per protocol. The demographic information of each of the 19

healthy Japanese male subjects who entered the study is presented in **Table 2**. One of three subjects withdrawn from the study was from the dose group of 20 mg/kg MK-3415A due to work commitments preventing from completion, and the other two subjects were from placebo group due to withdraw by himself or lost to follow-up.

**Table 2**. Summary of Demographic Data

| Treatment | Allocation Number | Age (years) | Height (cm) | Weight (kg) | BMI (kg/m$^2$) |
|---|---|---|---|---|---|
| Placebo | 0003 | 43 | 167.9 | 75.9 | 26.9 |
| | 0005 | 28 | 165.9 | 68.0 | 24.7 |
| | 0007 | 28 | 175.2 | 68.9 | 22.4 |
| | 0012 | 35 | 170.2 | 68.0 | 23.5 |
| | 0014 | 32 | 173.5 | 73.8 | 24.5 |
| | 0018 | 34 | 173.8 | 64.4 | 21.3 |
| 10 mg/kg MK-3415A | 0001 | 33 | 166.1 | 63.1 | 22.9 |
| | 0002 | 24 | 176.9 | 75.6 | 24.2 |
| | 0004 | 28 | 169.2 | 73.1 | 25.5 |
| | 0006 | 43 | 172.5 | 77.1 | 25.9 |
| | 0008 | 23 | 169.5 | 75.0 | 26.1 |
| | 0009 | 20 | 170.5 | 61.1 | 21.0 |
| 20 mg/kg MK-3415A | 0010 | 22 | 177.4 | 71.4 | 22.7 |
| | 0011 | 28 | 162.3 | 58.6 | 22.2 |
| | 0013 | 23 | 174.5 | 66.7 | 21.9 |
| | 0015 | 32 | 174.1 | 72.9 | 24.1 |
| | 0016 | 23 | 165.8 | 64.1 | 23.3 |
| | 0017 | 24 | 174.2 | 69.6 | 22.9 |
| | 0115 | 28 | 164.7 | 51.5 | 19.0 |
| **Study Summary:** | | | | | |
| Overall | N | 19 | 19 | 19 | 19 |
| | Range | 20-43 | 162.3-177.4 | 51.5-77.1 | 19.0-26.9 |
| | Mean | 29.0 | 170.75 | 68.36 | 23.42 |
| Placebo | N | 6 | 6 | 6 | 6 |
| | Range | 28-43 | 165.9-175.2 | 64.4-75.9 | 21.3-26.9 |
| | Mean | 33.3 | 171.08 | 69.83 | 23.88 |
| 10 mg/kg MK-3415A | N | 6 | 6 | 6 | 6 |
| | Range | 20-43 | 166.1-176.9 | 61.1-77.1 | 21.0-26.1 |
| | Mean | 28.5 | 170.78 | 70.83 | 24.27 |
| 20 mg/kg MK-3415A | N | 7 | 7 | 7 | 7 |
| | Range | 22-32 | 162.3-177.4 | 51.5-72.9 | 19.0-24.1 |
| | Mean | 25.7 | 170.43 | 72.9 | 22.30 |

**Pharmacokinetics of MK-6072:**

Mean MK-6072 concentrations versus time are presented for the 10 and 20 mg/kg MK-3415A groups on semi-logarithmic scale in **Figure 1**. Summary statistics for the MK-6072 PK parameters are presented in **Table 3**.

**Figure 1.** Mean (± Standard Deviation) Profiles for MK-6072 Serum Concentration Time Data (Semi-logarithmic Scale) (N = 6 for 10 mg/kg, N = 6 to 7 for 20 mg/kg)

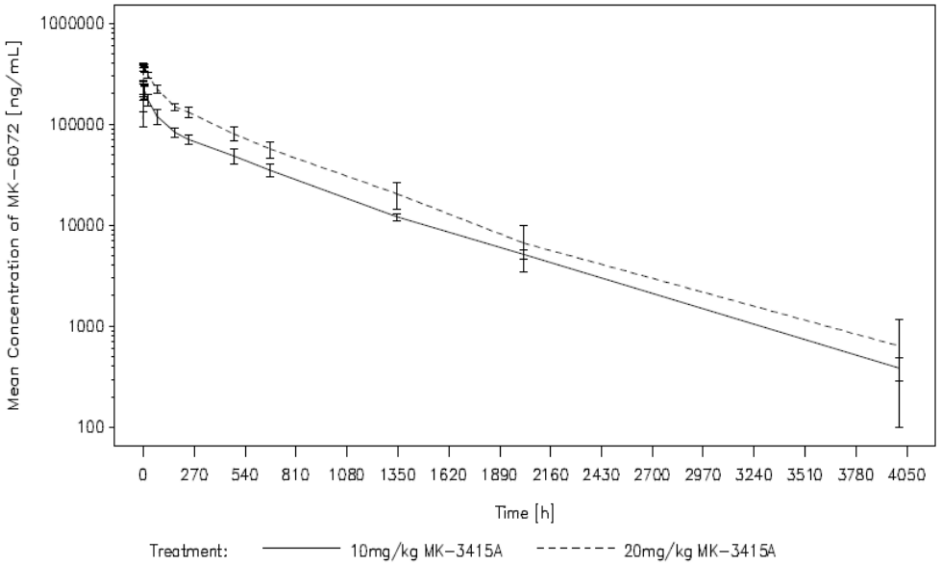



**Table 3**. Summary of Pharmacokinetic Parameters of MK-6072 by Treatment

| Analyte | Parameter | Statistic | 10 mg/kg MK-3415A (N=6) | 20 mg/kg MK-3415A (N=7) |
|---|---|---|---|---|
| MK-6072 | AUC0-∞ | n | 6 | 6 |
| | (h·µg/mL) | Arith.Mean | 73594 | 125099.5 |
| | | SD | 7505.47 | 18028.79 |
| | | CV(%) | 10.2 | 14.4 |
| | | Geo.Mean | 73284.9 | 124010.8 |
| | | Geo.CV(%) | 10.0 | 14.6 |
| | AUC0-last | n | 6 | 6 |
| | (h·µg/mL) | Arith.Mean | 73290.2 | 124645 |
| | | SD | 7495.3 | 17625.3 |
| | | CV(%) | 10.2 | 14.1 |
| | | Geo.Mean | 72980.2 | 123598.9 |
| | | Geo.CV(%) | 10.0 | 14.3 |
| | CL | n | 6 | 6 |
| | (mL/min) | Arith.Mean | 0.16168 | 0.17287 |
| | | SD | 0.014305 | 0.034612 |
| | | CV(%) | 8.8 | 20.0 |
| | | Geo.Mean | 0.16115 | 0.16978 |
| | | Geo.CV(%) | 8.9 | 21.5 |
| | Cmax | n | 6 | 7 |
| | (µg/mL) | Arith.Mean | 238.2 | 393.7 |
| | | SD | 35.63 | 21.99 |
| | | CV(%) | 15.0 | 5.6 |
| | | Geo.Mean | 235.9 | 393.2 |
| | | Geo.CV(%) | 15.4 | 5.7 |
| | t½ | n | 6 | 6 |
| | (h) | Geo.Mean | 530.44 | 481.87 |
| | | Geo.CV(%) | 7.1 | 16.9 |
| | Tmax | n | 6 | 7 |
| | (h) | Median | 1.020 | 1.020 |
| | | Min | 1.00 | 1.00 |
| | | Max | 2.00 | 8.00 |
| | Vz | n | 6 | 6 |
| | (L) | Arith.Mean | 7.42 | 7.15 |
| | | SD | 0.634 | 0.967 |
| | | CV(%) | 8.5 | 13.5 |
| | | Geo.Mean | 7.39 | 7.09 |
| | | Geo.CV(%) | 8.5 | 14.4 |

Arith.Mean = arithmetic mean; CV(%) = percent coefficient of variation; Geo.Mean = geometric mean;
Geo.CV% = geometric coefficient of variation; SD=standard deviation
Note: For Subject AN0015 (20 mg/kg MK-3415A group) all parameters except Cmax and Tmax were excluded from summary due to early discontinuation.
AUC0-∞, Vz and CL were calculated based on the predicted last measurable value.

When the 10 mg/kg MK-3415A data from the current study were compared to data obtained in a previous study of non-Japanese subjects (**Table 4**), the MK-6072 GMR [CI] of non-Japanese versus Japanese for $C_{max}$ was 1.08 [0.81, 1.44]. The MK-6072 GMR [CI] of non-Japanese versus Japanese for $AUC_{0-\infty}$ was 1.04 [0.88, 1.22] and for $AUC_{0-last}$ was 0.96 [0.80, 1.16]. The MK-6072 GMR [CI] of non-Japanese versus Japanese for CL was 1.08 [0.90, 1.30]. No significant difference in MK-6072 exposure was apparent between the ethnic groups.

**Table 4.** MK-6072 Geometric Mean Ratio of Ethnicity for Pharmacokinetic Parameters for 10 mg/kg Treatment (Pharmacokinetic Population)

| Parameter | Treatment | | Estimate | N | %CI | Lower CI | Upper CI |
|---|---|---|---|---|---|---|---|
| Analyte: MK-6072 | | | | | | | |
| AUC0-∞ | 10 mg/kg MK-3415A | Japanese | 73285 | 6 | 0.05 | 62571 | 85834 |
| (h·µg/mL) | | Non-Japanese | 76045 | 10 | 0.05 | 67282 | 85950 |
| | | Ratio Non-Japanese / Japanese | 1.038 | | 0.10 | 0.881 | 1.223 |
| AUC0-last | 10 mg/kg MK-3415A | Japanese | 72980 | 6 | 0.05 | 61317 | 86862 |
| (h·µg/mL) | | Non-Japanese | 70324 | 10 | 0.05 | 61450 | 80479 |
| | | Ratio Non-Japanese / Japanese | 0.964 | | 0.10 | 0.804 | 1.155 |
| CL | 10 mg/kg MK-3415A | Japanese | 0.161 | 6 | 0.05 | 0.135 | 0.192 |
| (mL/min) | | Non-Japanese | 0.174 | 10 | 0.05 | 0.152 | 0.200 |
| | | Ratio Non-Japanese / Japanese | 1.083 | | 0.10 | 0.903 | 1.298 |
| Cmax | 10 mg/kg MK-3415A | Japanese | 236 | 6 | 0.05 | 172 | 323 |
| (µg/mL) | | Non-Japanese | 256 | 29 | 0.05 | 222 | 295 |
| | | Ratio Non-Japanese / Japanese | 1.083 | | 0.10 | 0.813 | 1.443 |

CI = confidence interval; N = number of subjects
Note: Comparison between ethnic groups is based on study results (current Japanese study) and results of a second study (Non-Japanese, PN005, Cohort 1 for AUCs, Cohorts 1 and 2 for Cmax). Cohort 2 AUC data from PN005 were not used due to the truncated sampling time and unreliability of AUC data).
AUC0-∞ and CL were calculated based on the predicted last measurable value.

*Reviewer Comment:*

4) *PK parameters of MK-6072 in Japanese subjects are consistent with those in other ethnic groups. Thus, it can be reasonably concluded that there is no difference in PK of MK-6072 between Japanese and non-Japanese.*
5) *When administered in MK-3415A (a combination of MK-3415 and MK-6072), the half-life of MK-3415 is relatively longer than MK-6072 ($t_{1/2}$ of MK-3415 vs. MK-6072: ~30 vs. ~21 days), which is similar to the observation in Study P005 with non-Japanese subjects.*

**Immunogenicity:**

Serum was separated from blood samples and sent to a central laboratory for determination of ADA. Frequencies of the presence or absence of ADA by time point are presented in **Table 5**. All except one subject receiving MK-3415A returned negative results for the presence of ADAs. One subject in 20 mg/kg MK-3415A group returned a positive ADA result predose on Day 1. However, it was agreed between the principal investigator and the Sponsor that since the finding occurred predose and no further positive results were returned the event was not considered clinically relevant. No confirmatory ADA neutralizing test was performed.

**Table 5**. Frequencies of Presence of Anti-Drug Antibodies versus Time Point by Treatment

| Analyte | Visit | Time | Category | Placebo (N=6) n (%) | 10mg/kg MK-3415A (N=6) n (%) | 20mg/kg MK-3415A (N=7) n (%) |
|---|---|---|---|---|---|---|
| MK-3415 | Day 1 | Pre-dose | Absent | 6 (100.0) | 6 (100.0) | 5 ( 71.4) |
| | | | Present | 0 | 0 | 1 ( 14.3) |
| | Day 21 | 480 hours post-dose | Absent | 6 (100.0) | 6 (100.0) | 6 ( 85.7) |
| | Day 85 | 2016 hours post-dose | Absent | 6 (100.0) | 6 (100.0) | 6 ( 85.7) |
| | Day 168 | 4008 hours post-dose | Absent | 4 ( 66.7) | 6 (100.0) | 6 ( 85.7) |
| MK-6072 | Day 1 | Pre-dose | Absent | 6 (100.0) | 6 (100.0) | 6 ( 85.7) |
| | Day 21 | 480 hours post-dose | Absent | 6 (100.0) | 6 (100.0) | 6 ( 85.7) |
| | Day 85 | 2016 hours post-dose | Absent | 6 (100.0) | 6 (100.0) | 6 ( 85.7) |
| | Day 168 | 4008 hours post-dose | Absent | 4 ( 66.7) | 6 (100.0) | 6 ( 85.7) |

**Safety:**

All 19 enrolled subjects were included in the assessment of safety and tolerability. Nine (9 [47.4%]) subjects reported a total of 25 AEs; the majority of AEs were considered by the investigator to be mild in intensity, with one event (Subject AN0005: headache [placebo group]) considered moderate in intensity. All except 2 AEs were considered not related to study drug; one subject in 20 mg/kg MK-3415A group experienced 2 AEs (headache and arthralgia) considered to be possibly drug-related. In general, the incidence of AEs was equally distributed between the active dug treatment groups (5 events [10 mg/kg MK-3415A] versus 7 events [20 mg/kg MK-3415A]); no dose-related trend was observed. The highest incidence of AEs was observed in the placebo group. No AE required medical intervention apart from the administration of concomitant medication. There were no serious clinical, laboratory, or other AEs and no subjects died during the study.

**SPONSOR'S CONCLUSIONS:**

Safety Conclusions

- The 10 mg/kg MK-3415A and 20 mg/kg MK-3415A doses are equally safe and generally well tolerated in the Japanese male population.

Pharmacokinetic Conclusions

- Following a single intravenous infusion of 10 mg/kg and 20 mg/kg MK-3415A to healthy Japanese subjects Cmax and AUC parameters for MK-6072 in a slightly less than dose-proportional manner.
- Following a single intravenous infusion of 10 mg/kg and 20 mg/kg MK-3415A to healthy Japanese subjects CL for MK-6072 was consistent across dose levels, and between Japanese and non-Japanese subjects for the 10 mg/kg dose level.
- Elimination half-life appears to be independent of dose: approximately 20 days for MK-6072, consistent with the observation that clearance is not dose-dependent.
- Comparison of the 10 mg/kg MK-3415A data from the current study to data obtained in a previous study of non-Japanese subjects (PN005), showed that the pharmacokinetics of MK-6072 in healthy Japanese male subjects are generally similar to those observed in non-Japanese subjects.

Immunogenicity Conclusion

- No healthy Japanese male subject returned a positive anti-drug antibody (ADA) result at any time point after administration of MK-3415A (through day 168 post-infusion).

**REVIEWER ASSESSMENT:**
Following a single dose of MK-3415A (a combination of MK-3415 and MK-6072), the pharmacokinetics of MK-6072 in Japanese subjects are similar to those in non-Japanese subjects (Study P005), and the safety profile appears acceptable.

**Appendix 1**. Clinical Pharmacology Review for Meso Scale Discovery Sector Imager 6000 (MSD) Electrochemiluminescent (ECL) Method to Measure the Concentration of MK-6072 in Human Serum (Projects MYX, JBW2, JBW4, and JBW5)

Streptavidin-coated plates are blocked with 1% BSA/1XPBST blocking buffer for up to 3 hours at ambient room temperature. Calibrators (CALs) and quality controls (QCs), and samples all in 10% pooled human serum are loaded onto an intermediary assay plate or into cluster tubes. Biotinylated anti-15B7 and Ruthenylated anti-15B7 are diluted to a final concentration of 0.75 µg/mL in matrix buffer and added to all test wells of the intermediary assay plate or into the cluster tubes. The capture Ab, MK-6072, detection Ab complex is allowed to form during a 1 to 3 hour incubation period at room temperature. After the incubation, the blocking buffer is decanted from the Streptavidin coated plate. The labeled anti-drug-MK-6072 complex is loaded onto the blocked plate and incubated up to 1.5 hours at ambient room temperature. MK-6072 in the CALs, QCs and study samples is captured by the biotinylated anti-15B7 antibody and detected by the ruthenylated anti-15B7 antibody. Unbound materials are removed from the plate by washing and 1X MSD read buffer is added to the wells. MSD read buffer produces an electrochemiluminescent (ECL) signal, expressed in relative light units (RLU) that is proportional to the amount of MK-6072 bound by the capture and detection reagents. The conversion of RLU for the samples and the QCs to concentrations is achieved through a LIMS system computer software mediated comparison to a standard curve on the same plate, which is regressed according to a 5-parameter logistic (5-PL) regression model with a weighting factor of $1/Y^2$: $y = d + ((a - d) / (1 + (x / c)b)) (1/ratio^2)$. Results are reported in ng/mL concentration units.

**Sample Receipt**

Two hundred ninety-seven (297) human serum samples (and 309 replicates) were received frozen and in good condition (b) (4) between 08 November 2011 and 01 May 2012.

**Results**

Analysis of human serum samples began on 11 January 2012 and was completed on 01 June 2012. Each calibration curve was calculated using a five-parameter logistic ($1/response^2$ weighted) least-squares regression algorithm. Concentration data are reported to three significant figures. Back-calculated calibration standards are shown in **Table A1-1**. The average correlation coefficient for all obtained standard curves was 0.9983. Summary of Inter-assay Precision and Accuracy is shown in **Table A1-2**.

**Table A1-1**. Average Back-calculated Calibration Standards for Accepted Runs

| Run ID | CAL 1 (ng/mL) | CAL 2 (ng/mL) | CAL 3 (ng/mL) | CAL 4 (ng/mL) | CAL 5 (ng/mL) | CAL 6 (ng/mL) | CAL 7 (ng/mL) |
|---|---|---|---|---|---|---|---|
| N | [redacted] (b) (4) | | | | | | |
| Theoretical Concentration | | | | | | | |
| Mean | | | | | | | |
| S.D. | | | | | | | |
| %C.V. | | | | | | | |
| % Difference from Theoretical | | | | | | | |

**Table A1-2**. Statistical Summary of Inter-assay Precision and Accuracy for Accepted Runs

| Run ID | QC 2 Dil 10 (ng/mL) | QC 3 Dil 10 (ng/mL) | QC 4 Dil 10 (ng/mL) |
|---|---|---|---|
| N | 93 | 93 | 94 |
| Theoretical Concentration | 300 | 800 | 5120 |
| Mean | 305 | 784 | 5000 |
| S.D. | 18.6 | 45.5 | 296 |
| %C.V. | 6.11 | 5.81 | 5.93 |
| % Difference from Theoretical | 1.50 | -1.99 | -2.37 |
| Low Limit | 240 | 640 | 4100 |
| High Limit | 360 | 960 | 6140 |

No deviation from the method occurred, and no laboratory investigations were conducted, during the course of this study.

**Validation Results (Under Projects JBW2, JBW4, and JBW5)**

*(Please refer to Clinical Pharmacology Review for Study P005 for details)*

### 3.2. Pharmacometric Review

# 1 SUMMARY OF FINDNGS

The population pharmacokinetic (PK) model developed by the Applicant using data from three Phase 1 trials (PN004, PN005, and PN006) and two Phase 3 trials (PN001 and PN002), where MK-6072 was administered as single IV infusion alone or co-administered with MK-3415 (MK-2415A), is capable of characterizing the pharmacokinetics of MK-6072.

A two-compartment model with linear, first-order elimination best described the pharmacokinetics of MK-6072. Body weight was included in the model as a structural covariate on clearance (CL), intra-compartment clearance (Q), and central (Vc) and peripheral (Vp) volume of distribution. Albumin was found to be a major predictor of MK-6072 exposure as subjects with higher albumin levels associated with higher exposures. In addition to albumin, gender, race (black vs non-black), and Japanese race were also identified as significant covariates on CL or Vc.

The population PK estimates (%RSE) of MK-6072 were: CL 0.281 L/day (0.26%); apparent central volume of distribution (Vc) 3.43 L (0.79%); inter-compartmental clearance (CLp) 0.552 L/day (0.85%); peripheral volume of distribution (Vp) 3.57 L (1.14%); The inter-subject variability was 7.9% (28.7%) for CL and 1.1% (10.6%) for Vc.

An efficacy exposure-response (E-R) analysis was conducted using data from two Phase 3 studies in which MK-6072 was administered alone and in combination with MK-3415 with single dose of 10 mg/kg (note: MK-3415 was observed to have no clinical benefit compared to placebo on the endpoints under evaluation, and pooling of these treatment arms is considered to be reasonable). A significant reduction in *Clostridium difficile* infection (CDI) recurrence rate was observed after treatment with MK-6072 relative to placebo in the Phase 3 trials. A flat E-R relationship was identified between MK-6072 $AUC_{0\text{-}inf}$ and CDI recurrence suggesting that exposures achieved at the 10 mg/kg dose are on the maximal response plateau of the E-R curve. History of CDI in the past 6 months, baseline albumin level, age, and a Charlson Comorbidity Index≥3 were significant covariates affecting the rate of CDI recurrence in patients treated with both placebo and MK-6072.

In the analysis of all AEs during the four weeks following infusion, there was no association between the incidence of AEs and MK-6072 $AUC_{0\text{-}inf}$ ($p>0.05$). Covariates reflecting patient health, particularly concomitant medication use, were strong predictors of the incidence of AEs in the first four weeks following infusion. Concomitant use of non-SoC antibiotics or PPIs, race, and gender were significant covariates in patients who received placebo. An E-R analysis was also conducted for SAEs reported in the 12 weeks following infusion after treatment with MK-6072 or in combination with MK-3415. The analyses indicate that patient covariates, including albumin level and concomitant use of non-SoC antibiotics, rather than exposure, are the primary

factors influencing incidence of SAEs and that there is no clinically meaningful relationship between the incidence of SAEs in the 12 weeks following infusion and MK-6072 exposure.

## 1.1 Key Review Questions

The purpose of this review is to address the following key questions.

### 1.1.1 What covariates impact MK-6072 exposure based on the population PK model?

Body weight was a significant covariate on CL and Vc. At the 5th (45.5 kg) and 95th (109.6 kg) percentiles (P05 and P95) of the body weight distribution of the population PK dataset, the body weight effects result in a 18.5% decrease and 24.1% increase in clearance, respectively, relative to the clearance at the median body weight (69.7 kg). Similarly, Vc was 18% lower and 24% higher compared to Vc at the median body weight. Albumin was also a significant covariate on both CL and Vc. At the P05 (2.2 g/dL) and P95 (4.7 g/dL) of the albumin distribution of the population PK dataset, albumin effects result in a 47.7% increase and 25.2% decrease in CL, respectively, relative to the CL at the median albumin level (3.4 g/dL). Vc is 14.9% increased and 16.1% decreased compared to the Vc at the median albumin level. A typical male subject has 22% higher CL and 24% higher Vc compared to a typical female subject. Black race was associated with a 15% higher CL compared to a typical White subject. Japanese race was associated with a 9.5% lower CL and 14% lower Vc compared to a typical White subject. None of the effect of covariates was considered clinical relevant.

In addition, MK-6072 is eliminated by degradation through protein catabolism, and the pharmacokinetics is not expected to be substantially altered in patients with hepatic or renal impairment. The effect of hepatic or renal function on PK parameters was evaluated by exploratory analysis, further confirming that there was minimum impact. Therefore, the dose of 10 mg/kg appears to be appropriate approach for MK60-72. Dose adjustment is not necessary based on known intrinsic factors.

### 1.1.2 Are the PK parameters reported in the label supported by the Applicant's population PK?

Yes. Based on the Reviewer's analysis, the PK parameters derived from population PK model are summarized in Table 1. The results are comparable to the Applicant's PK parameters in the label.

**Table 1: Summary of MK-6072 PK parameter values in the patient population (n = 1515) Following Administration of a Single IV Dose of 10 mg/kg of MK-3415 or 10 mg/kg MK-3415 + MK-6072, based on Population**

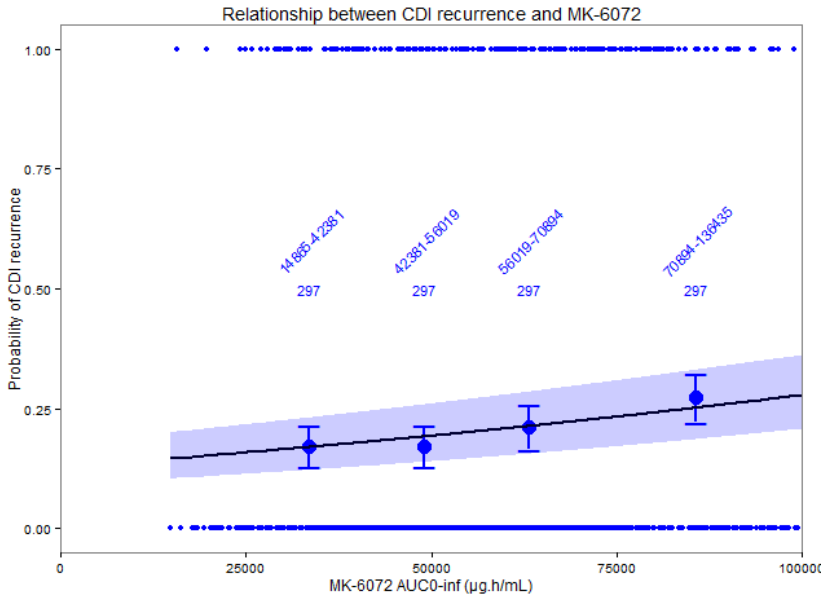
| | $AUC_{0\text{-}inf}$ (µg·h/mL) | $C_{max}$ (µg·h/mL) | t1/2 (day) | Tmax (h) | CL (L/day) | Vdss (L) |
|---|---|---|---|---|---|---|
| Geometric mean | 52604 | 178 | 18.7 | - | 0.317 | 7.33 |
| Geometric CV% | 40% | 44% | 28% | - | 41% | 16% |
| Median | 54285 | 189 | 18.9 | 1.33 | 0.310 | 7.36 |

*Source: Reviewer's analysis*

### 1.1.3 What are the characteristics of E-R relationship for efficacy? Does it support the proposed dose regimen?

A significant relationship was identified between exposure and CDI recurrence with higher exposure associated with a higher CDI (Figure 1). The logistic regression analysis showed that in addition to exposure, several other patient covariates also had a significant impact on the probability of CDI recurrence, including baseline albumin (note: albumin lab values were only available at baseline and week 4), history of CDI in the past 6 months, age, and a Charlson Comorbidity Index ≥ 3. Since albumin is highly correlated with MK-6072 exposure (AUC) and also associated with the patient health, the relationship between CDI recurrence and MK-6072 exposure is confounded with baseline albumin.

**Figure 1: Relationship between CDI recurrence and MK-6072**




*The number with ranges on the top represent the MK-6072 exposure ranges for each bin and the numbers under the ranges represent the sizes of each bin.*

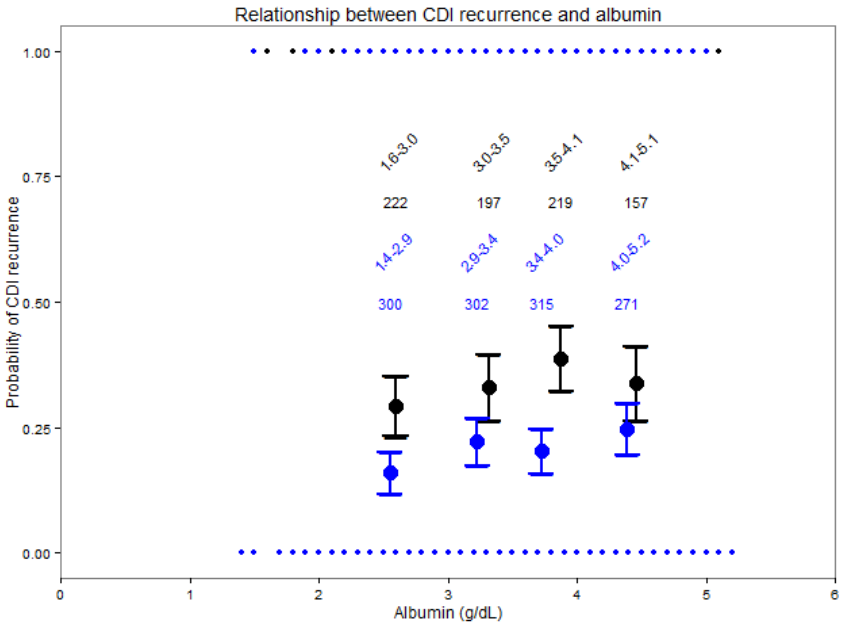
*Note: The reviewer used dataset excluding the patients who experienced clinical initial failure during standard of care.*

*Source: Reviewer's analysis*

To better understand the relationship between albumin and CDI recurrence, the Reviewer plotted the CDI recurrence versus four quartiles of baseline albumin as shown in Figure 2 for both treatment and placebo. Overall, CDI recurrence increases with higher albumin concentrations in both treatment arms. One advantage of looking at albumin in this manner is that it permits a direct quartile-to-quartile comparison of the odds ratio between the treatment and placebo arms. While an overall trend in response with respect to albumin may be observed within a treatment arm, if the same trend is also observed in the placebo arm this suggests it is

patient characteristics that are contributing factors to the relationship and not MK-6072 exposure. As shown in Table 2, the odds ratio between placebo and treatment are relatively consistent across all quartiles, supporting a consistent treatment effect across all quartiles. The results suggest that the exposure provided by 10 mg/kg MK-6072 approach a flat response rate regarding CDI recurrence and that higher exposure is not predicted to produce further benefit. Therefore, no dose adjustment is necessary based on this observation.

**Figure 2: Relationship between CDI recurrence and albumin**




*Blue depicts the treatment arm and black depicts the placebo arm. The numbers with ranges on the top represent the albumin concentration ranges for each bin and the numbers under the ranges represent the size of each bin.*

*Source: Reviewer's analysis*

**Table 2: Odds ratio of CDI recurrence for the treatment arm compared to the placebo arm across the four quantiles of albumin**

| Albumin quantile | 1st quantile | 2nd quantile | 3rd quantile | 4th quantile |
|---|---|---|---|---|
| Odds ratio | 0.55 | 0.67 | 0.52 | 0.73 |

*Source: Reviewer's analysis*

### 1.1.4 What are the characteristics of E-R relationship for safety? Does it support the proposed dose regimen?

There were no apparent differences in the overall adverse event rate, severe adverse event rate, or types of adverse events observed between placebo and the treatment arms. In total 61.7% and 61.2% of subjects reported one or more adverse events in the treatment and placebo

arms, respectively. Serious adverse events were slightly more frequent in the placebo arm (32.7%) compared to the treatment arm (29.4%). The most frequent serious adverse events were gastrointestinal disorders and infections and infestations (Table 3). Adverse events at these sites could include diarrhea, which as discussed above, was one of the components of the efficacy analysis. As such, the imbalance in serious adverse events favoring treatment may reflect treatment effect in reducing the occurrence of diarrhea associated with CDI.

**Table 3: Subjects with Serious Adverse Events (>3%) During 12 Weeks Following Infusion Phase 3 Studies (P001, P002, and P001 + P002 Integrated)**

| | (Pooled) MK-3415A | (Pooled) MK-6072 | (Pooled) Placebo |
|---|---|---|---|
| Total subjects No. | 777 | 786 | 781 |
| With one or more serious adverse events | 212 (27.3%) | 231 (29.4%) | 255 (32.7%) |
| Cardiac disorders | 24 (3.1%) | 36 (4.6%) | 27 (3.5%) |
| Gastrointestinal disorders | 37 (4.8%) | 49 (6.2%) | 42 (5.4%) |
| Infections and infestations | 93 (12.0%) | 104 (13.2%) | 138 (17.7%) |
| Nervous system disorders | 24 (3.1%) | 13 (1.7%) | 8 (1.0%) |
| Respiratory, thoracic and mediastinal disorders | 26 (3.3%) | 28 (3.6%) | 24 (3.1%) |

*Source: Applicant's summary of clinical safety report, Page 377, Appendix 2.7.4: 16 (Adapted)*

Based on the reported AEs and SAEs from treatment and placebo arms, comparable incidence of AE or SAE is observed between treatment and placebo arms, and thus no significant E-R relationship is identified.

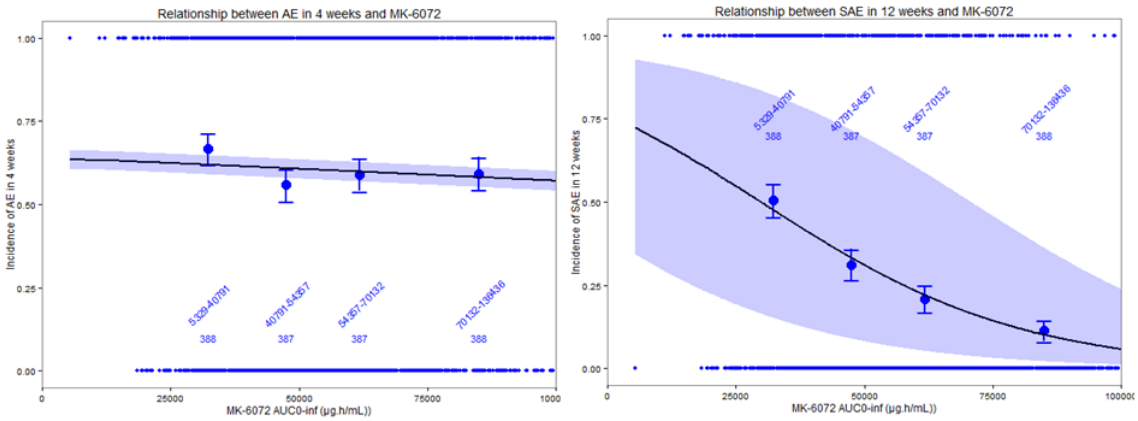
The reviewer also conducted an additional sensitivity analysis to evaluate the difference between treatment arm and placebo arm by quantile of albumin, the result is in line with the conclusion.

There was no relationship identified between MK-6072 exposure and AEs in 4 weeks, while there was a significant relationship identified between MK-6072 exposure and SAE in 12 weeks (Figure 3). For SAE in 12 weeks, the relationship indicated that higher exposure would result in lower severe safety events. The logistic regression analysis shows that in addition to exposure, several patient covariates were significantly associated with SAE in 12 weeks, including albumin, concomitant use of non-SoC systemic antibiotics or PPIs, hospitalization, Charlson Comorbidity Index ≥ 3, and age. As albumin is confounded with MK-6072 exposure and patient health, the incidence of SAE in 12 weeks was plotted against four quantiles of albumin concentrations at baseline (Figure 4) and odds ratios were compared between treatment arm and placebo arm (Table 4). The rationale for this analysis is similar to that outlined in the exposure-response efficacy analyses above.

The odds ratios are relatively flat across albumin quartiles and treatment arm always had lower SAEs compared to placebo arm. A possible reason for this observation is that the AEs included

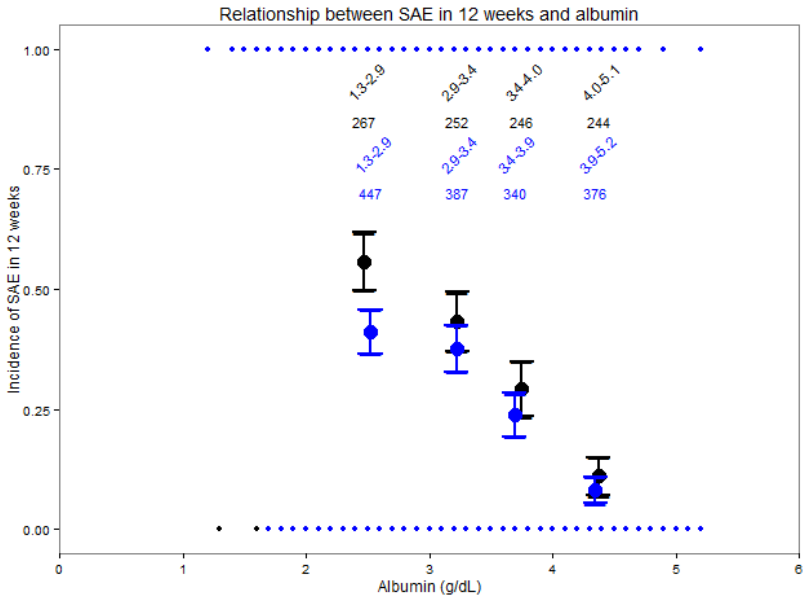
diarrhea, which is also a component of the efficacy endpoint (and associated with drug effect). Overall, the results indicate that no dose adjustment is necessary based on safety observations.

**Figure 3: Relationship between incidence of AEs in 4 weeks or SAEs in 12 weeks and MK-6072**




*The numbers with ranges on the top or in the middle represent the MK-6072 exposure ranges for each bin and the numbers under the ranges represent the size of each bin.*
*Source: Reviewer's analysis*

**Figure 4: Relationship between incidence of SAEs in 12 weeks and albumin**




*Blue depicts the treatment arm and black depicts the placebo arm. The numbers with ranges in the middle or on the top represent the albumin concentration ranges for each bin and the numbers under the ranges represent the size of each bin.*
*Source: Reviewer's analysis*

**Table 4: Odds ratios of treatment arm to placebo arm across the four quartiles of albumin for SAE in 12 weeks**

| Albumin quartile | 1st quartile | 2nd quartile | 3rd quartile | 4th quartile |
|---|---|---|---|---|
| SAE in 12 weeks | 0.74 | 0.87 | 0.81 | 0.75 |

*Source: Reviewer's analysis*

## 1.2 Recommendations

The Division of Pharmacometrics (Office of Clinical Pharmacology) has reviewed this application and recommends approval of a single IV infusion administration of MK-6072 (10 mg/kg). The reviewer agrees with the Applicant's conclusion from the population PK analysis that no dose adjustments are necessary for MK-6072 based on age, body weight, sex, race, renal function, hepatic impairment in adult patients. In addition, the exposure-response relationship for efficacy and safety support the proposed dose of 10 mg/kg MK-6072 single infusion administration in adult patients.

# 2 RESULT OF APPLICANT'S ANALYSES

The pharmacometric analyses covered in this review are the Applicant's population PK and exposure-response analyses.

## 2.1 Population PK Model

### 2.1.1 Objectives

- To evaluate the impact of intrinsic and extrinsic factors by stepwise covariate model (SCM). Intrinsic factors of particular interest are body weight, gender, age, race, and renal function. Extrinsic factors focused on potential DDIs with common concomitant medication and between MK-3415 and MK-6072.
- To qualify the final model by visual predictive check (VPC).
- To evaluate the effect of covariates in the final population PK model by simulation, where the univariate effect of each covariate was considered with all other covariates held constant at typical patient values.

### 2.1.2 Trial included in the population PK model

The population PK analysis included data from three Phase 1 trials and two Phase 3 trials, summarized in Table 5 and 6. While the dense PK sampling scheme in the Phase 3 trials was expected to be sufficient to adequately characterize the pharmacokinetics of MK-6072, the Phase 1 trials were also included in the analysis as they contain data from Japanese subjects and treatment with the 20 mg/kg dose.

**Table 5: Phase 1 Trials Included in the Population PK Model**

| Protocol number | Design | Dose (mg/kg) | Endpoints | Actual Sample Size | PK sampling[1] |
|---|---|---|---|---|---|
| PN004 | Healthy subjects, two sequential doses | 10 mg/kg MK-3415 + 10 mg/kg MK-6072, repeated after 12 wks | PK, safety, ADA | 30 | Pre-dose, 0.5, 1, 2, 4, 8 hr and 2, 8 and 43 days post first dose . Predose on day 85 and then 0.5, 1,2,4,8 hr post-dose and then days 86, 92, 127, 169 and 253 |
| PN005 | Healthy subjects, single dose | 10 mg/kg MK-3415 + 10 mg/kg MK-6072 | PK, safety | 29 (10A+10B) 6 placebo | Pre-dose, 0.5, 1, 2, 4, 6, 8, 24, 72 hr and 7, 10, 20, 28, 56 and 84 days post dose |
| PN006 | Japanese healthy subjects, single dose | 10 mg/kg MK-3415 + 10 mg/kg MK-6072, 20 mg/kg MK-3415 + 20 mg/kg MK-6072, Placebo | PK, safety | 6 (10A+10B) 7 (20A+20B) 6 placebo | Pre-dose, 0.5, 1, 2, 4, 6, 8, 24, 72 hr and 7, 10, 20, 28, 56, 84 and 168 days post dose |

*Source: Applicant's modeling and simulation report, Page 44, Table 2.*

**Table 6: Phase 3 Trials Included in the Population PK and Exposure-Response Models (Per Protocol)**

| Protocol number | Design | Dose (mg/kg) | End points | Intended Sample Size | PK sampling[1] |
|---|---|---|---|---|---|
| PN001 | Patients with CDI, adaptive design, single dose adjunctive to SOC | 10 mg/kg MK-3415, 10 mg/kg MK-6072, 10 mg/kg MK-3415 + 10 mg/kg MK-6072 (MK-3415A), placebo | Efficacy (Proportion of patients with CDI recurrence), Safety, PK, ADA | 1450 (n=400 per MK-6072, MK-3415A and placebo arms, 250 per MK-3415 arm) | PK: Pre-dose, 1 hr post infusion and 3, 10, 28, 56 and 84 days post dose, also unscheduled if CDI recurs ADA: predose and 28, 56 and 84 days post dose |
| PN002 | Patients with CDI, single dose adjunctive to SOC | 10 mg/kg MK-6072, 10 mg/kg MK-3415 + 10 mg/kg MK-6072 (MK-3415A), placebo | Efficacy (Proportion of patients with CDI recurrence), Safety, PK, ADA | 1200 (n=400 per treatment arm) | PK: Pre-dose, 1 hr post infusion and 3, 10, 28, 56 and 84 days post dose, also unscheduled if CDI recurs ADA: predose and 28, 56 and 84 days post dose |

*Source: Applicant's modeling and simulation report, Page 45, Table 3.*

### 2.1.3 Data analysis plan

The population PK modeling was performed with the software package NONMEM, version VII (Globomax, 7250 Parkway Drive, Suite 430, Hanover, MD 21076 USA) running under PsN (Perl-speaks-NONMEM) 3.4.2. Model fitting was performed in a UNIX environment with Intel FORTRAN Compiler, version 11.1 (Intel Corporation, 2200 Mission College Blvd., Santa Clara, CA 95054). Post processing of the data was conducted using R 2.15.2(The R Foundation for Statistical Computing, Vienna, Austria).

The covariates investigated in the population PK model are listed in Table 7.

**Table 7: Covariates included in the Population PK or Exposure-Response Analysis**

| Covariate | Classification / derivation | Included in popPK Analysis | Included in E-R Analysis |
|---|---|---|---|
| Treatment | Categorical: 0=Placebo; 1=MK-3415 alone; 2=MK-6072 alone; 3=MK-3415A | | X |
| MK-3415 presence | Categorical: 0=Placebo and MK-6072 alone treatment; 1=MK-3415 alone and MK-3415A treatment | X | |
| MK-6072 presence | Categorical: 0=Placebo and MK-3415 alone treatment; 1=MK-6072 alone and MK-3415A treatment | X | |
| Age (years) | Continuous | X | X |
| Weight (kg) | Continuous | X | X |
| BMI ($kg/m^2$) | Continuous | | X |
| Gender | Categorical: 0=male; 1=female | X | X |
| Race | Categorical, re-grouped: 0=White or Other; 1=Black/African American; 2=Asian | X | X |
| Ethnicity | Categorical: 0=non-Hispanic; 1=Hispanic | X | |
| Japanese | Categorical: 0=non-Japanese race; 1=Japanese race | X | |
| Modification of Diet in Renal Disease estimated glomerular filtration rate (MDRD eGFR) ($mL/min/1.73\ m^2$) | Continuous<br>eGFR for women = 175 *(SCr^-1.154) * (Age^-0.203) * 0.742<br>eGFR for men = 175 *(SCr^-1.154) * (Age^-0.203)<br>Multiple by 1.212 if African American | X | |
| Patient status | Categorical: 1=Patient (P001, P002); 2=Healthy Subject (P004, P005, P006) | X | |
| Hospitalization Status | Categorical: 0=HV or Outpatient; 1=Inpatient | Exploratory | X |
| Modified Horn's index | Categorical, re-grouped: 0=HV or MHIX score 1 (low) or 2 (moderate); 1=MHIX score 3 (severe) or 4 (extreme) | | X |
| Charlson Comorbidity Index | Categorical, re-grouped: 0 = HV or < 3; 1 = ≥ 3 | X | X |
| Clinically Severe CDI (Zar score ≥ 2) | Categorical, re-grouped: 0=HV or Zar score < 2; 1=Zar score ≥ 2 | Exploratory | X |
| Antidrug antibody MK-3415 | Categorical: 0 = Negative; 1 = Inconclusive; 2 = Non- Treatment emergent positive; 3 = Treatment emergent positive; 4=potential positive; 5= no meaningful pre or post dose results | Exploratory | |
| Antidrug antibody MK-6072 | Categorical: 0 = Negative; 1 = Inconclusive; 2 = Non- Treatment emergent positive; 3 = Treatment emergent positive; 4=potential positive; 5= no meaningful pre or post dose results | Exploratory | |
| Albumin (g/dL) | Continuous | X | X |
| Hepatic Impairment | Categorical: 0 = No or HV; 1=Yes | X | |
| Endogenous IgG for anti-toxin A | Categorical, re-grouped: -1=HV; 0=<1:1000 or 1:1000; 1=1:5000 or 1:25000 or 1:125000 or 1:125,000 | | X |
| Endogenous IgG for anti-toxin B | Categorical, re-grouped: -1=HV; 0=<1:1000 or 1:1000; 1=1:5000 or 1:25000 or 1:125000 or 1:125,000 | | X |
| Baseline white blood cell count ($10^3/uL$) | Continuous | | X |
| Compromised Immunity | Categorical: 0=No or HV; 1=Yes; .=Unknown | | X |
| History of CDI in past 6 months | Categorical: 1=Yes; 2=No or HV; 3=Unknown | | X |
| 027 Ribotype | Categorical, 0=027 Ribotype; 1 = other ribotype; -1=HV; .=Unknown | | Exploratory[†] |
| Standard of care medication | 1=metronidazole, 2 = vancomycin, 3 = fidaxomycin ; -1= HV | X | X |
| Concomitant non-SoC antibiotic use | Categorical, 0=No or HV; 1=Yes | X | X |
| Concomitant PPI use | Categorical, 0=No or HV; 1=Yes | X | X |

X: included as covariate in the covariate analysis; Exploratory: only used for exploratory analysis; Scr: serum creatinine (mg/dL); PPI: proton pump inhibitors

[†]Ribotype was not included in the covariate analysis as > 30% of the population was "Unknown"

*Source: Applicant's modeling and simulation report, Page 49, Table 4.*

### 2.1.4 Structural model building

Model development was carried out using first-order, conditional estimation with interaction (FOCEI). Models were developed in order of increasing complexity, beginning with very simple models (e.g. 1-compartment with 1st-order elimination) and proceeded no further improvement in fit was supported by the data. As dosing of MK-6072 is weight-based, weight was considered a structural covariate, which was included early in model development prior to full covariate evaluation.

The inter-individual random effects were mostly assumed to be independently distributed, i.e. the corresponding off-diagonal component of Ω was assumed to be 0. However, correlations in Ω were checked graphically using a pairs plot, and if plausible correlations were found (e.g. between CL and Vc), their significance in the model was checked by estimating the corresponding off-diagonal element(s).

The goodness-of-fit and appropriateness of the included random effects was assessed by means of diagnostic plots such as: histogram of ETA estimates, co-plots of individual ETA estimates, (absolute) individual weighted residuals versus individual predictions, and histograms of population and individual weighted residuals. In addition, shrinkage estimates were calculated for all IIV estimates and precision of the IIV estimate was also considered.

### 2.1.5 Covariate analysis

A full covariate search was performed on the base model to evaluate the impact of covariates on PK. An automated forward inclusion followed by backward elimination procedure was followed through use of the step-wise covariate modeling procedure (SCM) tool as implemented in PsN. This procedure involved stepwise testing of covariate relationships in a forward inclusion (ΔOFV of 3.84, $p < 0.05$ for 1 DF) and backward exclusion (ΔOFV of 7.88, $p < 0.005$ for 1 DF) procedure. For categorical covariates, the ΔOFV for the respective p-values may be different depending on the degrees of freedom.

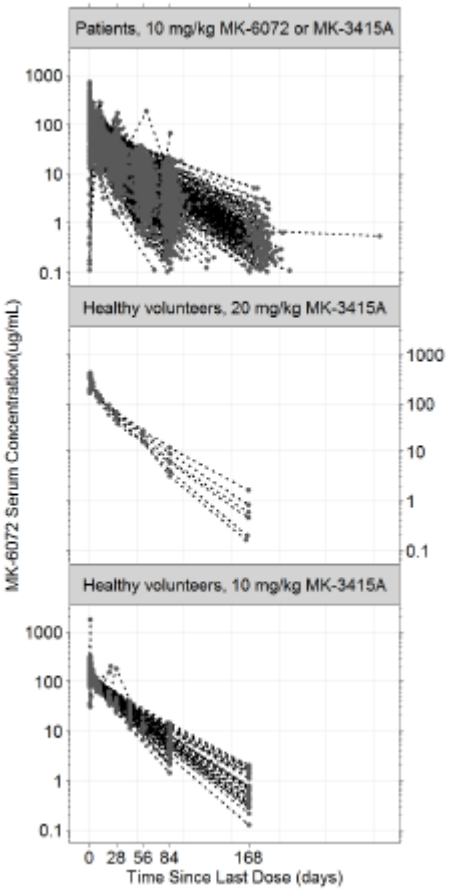
Significant covariates with marginal effects identified in the SCM were further evaluated for improvement of model fit. Reductions in ETA and residual variability, shifts in ETA distributions and diagnostics plots with and without the covariates were investigated. Rejection of marginal-effect covariates was based on small effect sizes on CL and Vc (i.e. <10%), relative standard errors >25%, and lack of improvement in model fit and diagnostics.

### 2.1.6 Population PK results

Figure 5 shows MK-6072 serum concentrations over time (time after first dose) stratified by dose for Phase 1 and Phase 3 trials, respectively. Profiles of both compounds appear to decline in a biphasic manner, indicating a two-compartmental model might be suitable to describe the pharmacokinetics of MK-6072. There was no evidence of target mediated drug disposition (TMDD) based on visual inspection of individual concentration-time profiles.

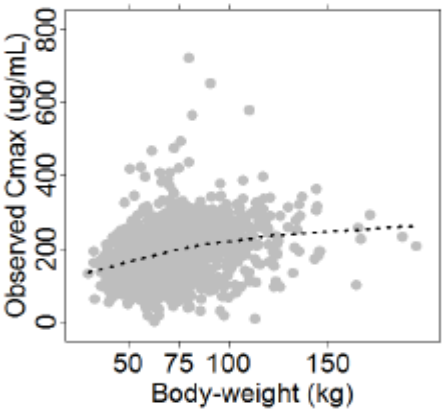
The correlation plots of MK-6072 $C_{max}$ versus body weight presented in (Figure 6) show a tendency of increasing $C_{max}$ with increasing body weight ($C_{max}$ increasing by 35% from a body weight of 50 to 100 kg).

**Figure 5: MK-6072 serum concentration-time plots for Phase 1 and Phase 3 trials**




*Source: Applicant's modeling and simulation report, Page 77, Figure 3.*

**Figure 6: Relationship between Observed MK-6072 $C_{max}$ and Body weight**




*Source: Applicant's modeling and simulation report, Page 78, Figure 4.*

### *2.1.6.1 Structural model selection*

A two-compartment model with an estimated allometric scaling for weight on all PK parameters was selected as the structural model to describe the serum concentration-time profiles for MK-6072. The base model for MK-6072 included ETAs on CL and Vc. Although addition of an ETA on Vp resulted in a significant decrease in OFV, it was not retained in the model because of high shrinkage (55%).

Multiple residual variability models were also explored, including log-additive and combined log-additive and additive models. The combined log-additive and additive residual error model resulted in a significant decrease in OFV compared to the log-additive error model. However, this model appeared to be numerically less stable. Consequently, the log-additive residual error model was selected for the MK-6072 population PK models.

The base models described the observed serum concentration data well. Parameter estimates of the base models are shown in Table 8.

**Table 8: Parameters Estimates of the Base Model for MK-6072**

| **Parameters** | **Estimates** | **% RSE[e]** | |
|---|---|---|---|
| CL(L/d) | 0.310 | 0.29% | |
| Vc (L) | 3.69 | 0.83% | |
| Q (L/d) | 0.553 | 0.84% | |
| Vp (L) | 3.60 | 1.2% | |
| $\alpha^{d}$ for CL, Q[a] | 0.482 | 8.2% | |
| $\alpha^{d}$ for Vc, Vp[b] | 0.517 | 5.6% | |
| **Random Effect** | **Estimates (%CV)[c]** | **%RSE[e]** | **Shrinkage** |
| IIV on CL | 37.6% | 6.2% | 0.9% |
| IIV on Vc | 18.9% | 19.1% | 9.8% |
| IIV on Residual Error | 59.5% | 9.7% | -3.5% |
| **Residual Error** | **Estimates** | **% RSE[e]** | |
| Log-additive | 0.177 | 2.8% | 5.3% |

[a]CL (Q)i =CL (Q)I x (WGTi/median(WGT))$^{0.482}$ ; [b]Vi=V x (WGTi/median(WGT))$^{0\ 517}$ ; [c]Percentage of coefficient of variation (%CV) calculated as sqrt((exp(Omega)-1)x100; [d]α : power value for weight-based scaling; [e]%RSE derived from bootstrap analysis, calculated as 100*standard error/median

*Source: Applicant's modeling and simulation report, Page 85, Table 15.*

### *2.1.6.2 Final model selection*

Overall, 17 covariates were evaluated on CL and Vc of MK-6072 during the formal covariate testing via the SCM algorithm. The covariates included from the forward selection step for both

PK models were tested against the backwards selection criteria (p <0.005) and non-significant covariates were removed one by one according to the SCM algorithm.

Parameter estimates from the final model and relative standard errors and confidence intervals obtained from bootstrap analysis of the final models are presented in Tables 9 and 10.

**Table 9: Parameter Estimates of Structural Components of Final Population PK Models**

| | MK-6072 | | |
|---|---|---|---|
| **Parameters** | **Estimate** | **%RSE [c]** | **95% CI [a]** |
| CL (L/day) | 0.281 | 0.257 | 0.275 ; 0.288 |
| Vc (L) | 3.43 | 0.793 | 3.37 ; 3.50 |
| Q (L/day) | 0.552 | 0.851 | 0.518 ; 0.588 |
| Vp (L) | 3.57 | 1.14 | 3.48 ; 3.68 |
| [b]α for CL and Q | 0.477 | 7.04 | 0.409 ; 0.542 |
| [b]α for Vc or Vp | 0.477 | 5.11 | 0.426 ; 0.524 |
| **Covariates on CL** | | | |
| Albumin (power) | -0.897 | 4.07 | -0.968 ; -0.825 |
| Japanese | -0.0947 | 28.8 | -0.141 ; -0.0347 |
| Race - Black | 0.154 | 25.1 | 0.0801 ; 0.231 |
| Gender - Male | 0.219 | 9.56 | 0.18 ; 0.261 |
| **Covariates on Vc** | | | |
| Albumin (linear) | -0.124 | 7.03 | -0.141 ; -0.107 |
| Japanese | -0.144 | 20.1 | -0.195 ; -0.0846 |
| Race - Asian | -- | -- | -- |
| Gender - Male | 0.243 | 7.23 | 0.209 ; 0.277 |

*Source: Applicant's modeling and simulation report, Page 120, Table 24.*

**Table 10: Parameter Estimates of Stochastic Components Final Population PK Models**

| **Random** | **MK-6072** | | |
|---|---|---|---|
| **Effect** | **Estimates** | **95% CI [a]** | **Shrinkage** |
| IIV[b] CL | 0.0791 | 0.0717 ; 0.0867 | |
| | (28.7%)[c] | (27.3 ; 30.1) | 3.1% |
| IIV Vc | 0.0111 | 0.0061 ; 0.0164 | |
| | (10.6%) | (7.8 ; 12.9) | 26.1 |
| IIV Epsilon | 0.328 | 0.286 ; 0.384 | |
| | (62.3%) | (57.5 ; 68.4) | -1.1% |
| **Residual** | | | |
| **Error** | **Estimates** | **95% CI** | **Shrinkage** |
| Log-add. | 0.182 | 0.174 ; 0.191 | 3.9% |

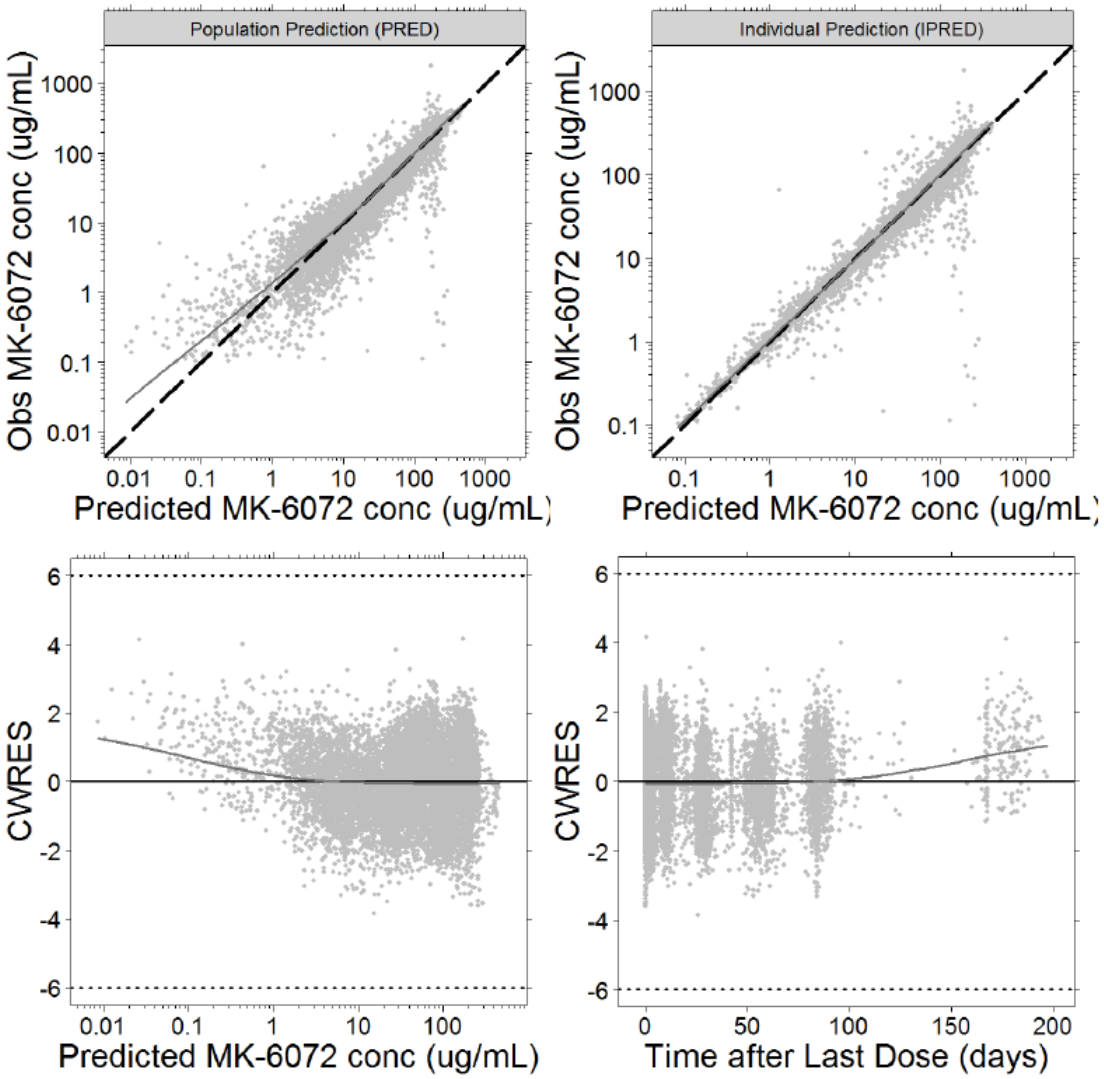
*Source: Applicant's modeling and simulation report, Page 121, Table 25.*

### 2.1.7 Evaluation of diagnostics for the final model

The first row of Figure 7 show scatter plots of individual and population predicted concentrations vs. observed concentrations. In the figure the loess smooth line of the data points and the line of identity are in good agreement. The bias observed in the lower concentration range for the base models is reduced in the final models. Overall, the figure indicates an adequate fit of the MK-6072 models to the data.

Plots of conditional weighted residuals versus population predicted values and conditional weighted residuals versus time after dose are presented in the lower panels of Figure 7. Although some bias is observed in the concentration range near LLOQ, the vast majority of the data is well predicted and there are no systematic trends in the clinically relevant concentration range and time range (Day 1 – 85).

**Figure 7: Goodness of Fit Plots for the Final MK-6072 Population PK model**



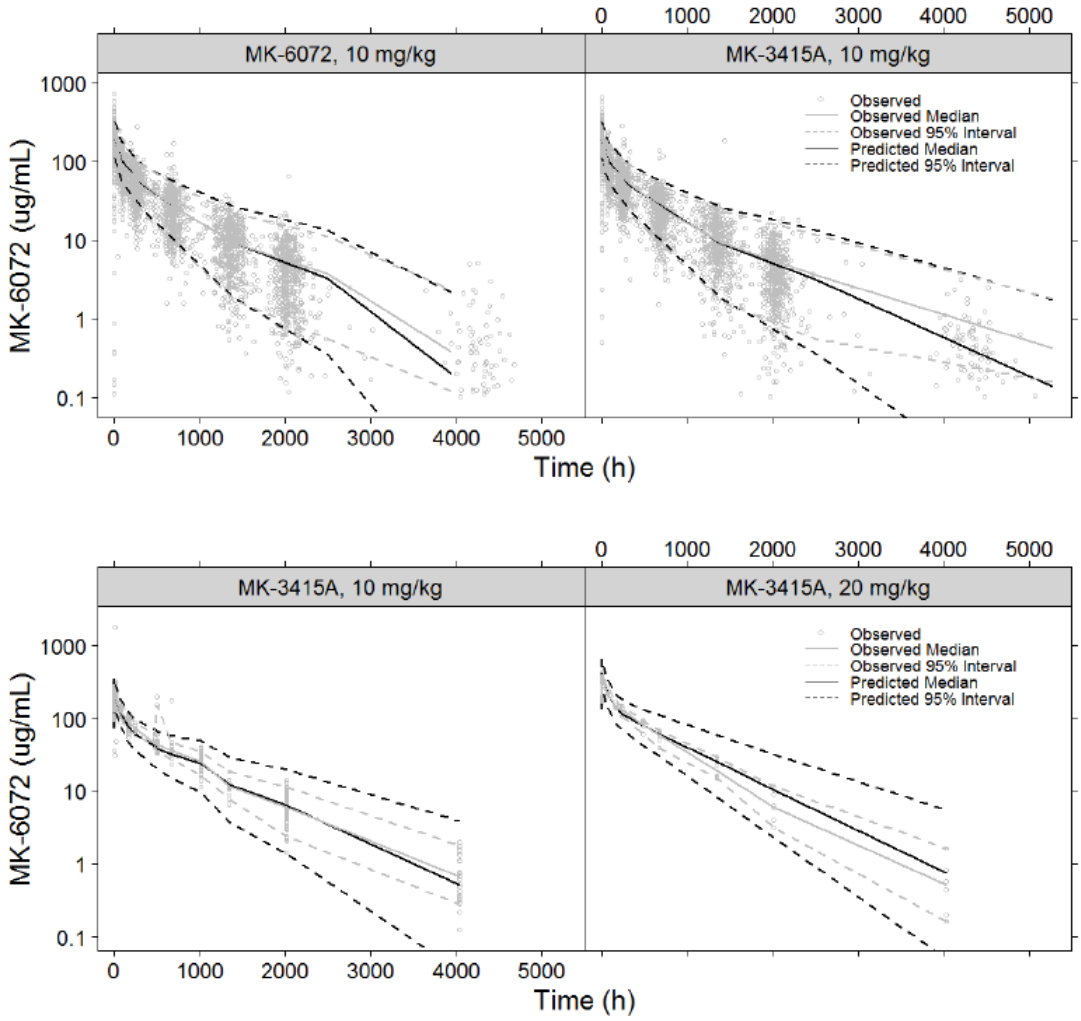

*Source: Applicant's modeling and simulation report, Page 122, Figure 52.*

### 2.1.8 Visual predictive check (VPC)

Visual predictive checks stratified by population (healthy subject, patient) and treatment (10 mg/kg MK-3415A, 10 mg/kg MK-6072, and 20 mg/kg MK-3415A) were performed. Figure 8 presents the VPC for MK-6072 serum concentrations in patients and healthy subjects upon treatment with a single dose of either MK-6072 or MK-3415A.

The final models of MK-6072 were able to predict the observed median and 5th and 95th percentile of observed MK-6072 and MK-3415 concentrations with good accuracy.

**Figure 8: Visual Predictive Check for MK-6072 Serum Concentrations from P001 and P002 (upper plots) and P004, P005, P006 (lower plots), Stratified by Treatments**




*Source: Applicant's modeling and simulation report, Page 133, Figure 62.*

### 2.1.9 Assessment of the magnitude of introduced covariate effects on PK parameters

The covariate relationship included in the final models for the continuous covariates body weight and albumin were simulated 1000 times based on uncertainty in the parameter

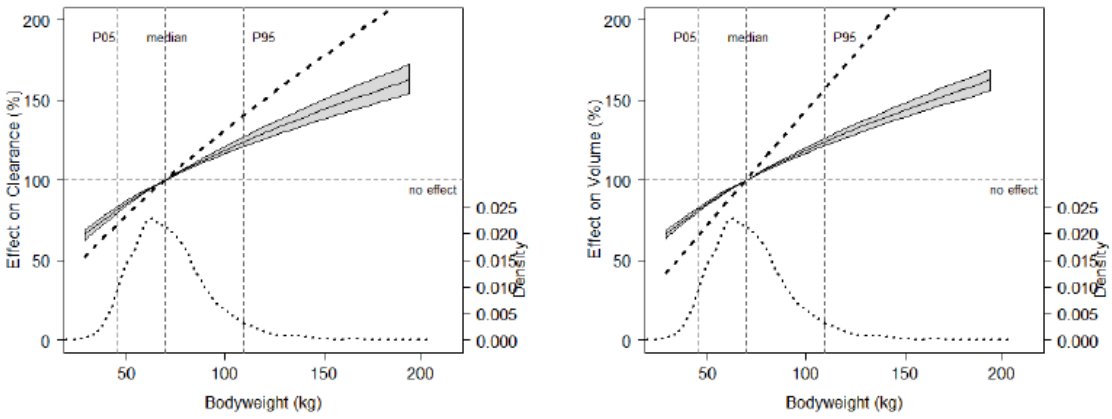
estimates to characterize the effect of these covariates on clearance and central volume of distribution.

### *2.1.9.1Assessment of the magnitude of body weight as covariate on clearance and central volume of distribution*

Body weight was included as a power function on clearance and volume parameters of the MK-6072 population PK models. Allometric coefficients for MK-6072 clearance and volume parameters in the final model were 0.477. The thick dashed lines in Figure 9 represent the body weight relationship assuming allometric scaling coefficients of 0.75 and 1 for clearance and volume parameters, respectively.

At the 5th (45.5 kg) and 95th (109.6 kg) percentiles (P05 and P95) of the body weight distribution of the population PK dataset, the body weight effects result in a 18.5% decrease and 24.1% increase in clearance, respectively, relative to the clearance at the median body weight (69.7 kg). Similarly, central volume is 18.4% decreased and 24.1% increased at the P05 and P95 of the body weight distribution as compared to the central volume at the median body weight.

**Figure 9: Distribution of observed Body Weight and included Body Weight effect on the typical value of MK-6072 clearance (left) and central volume (right)**

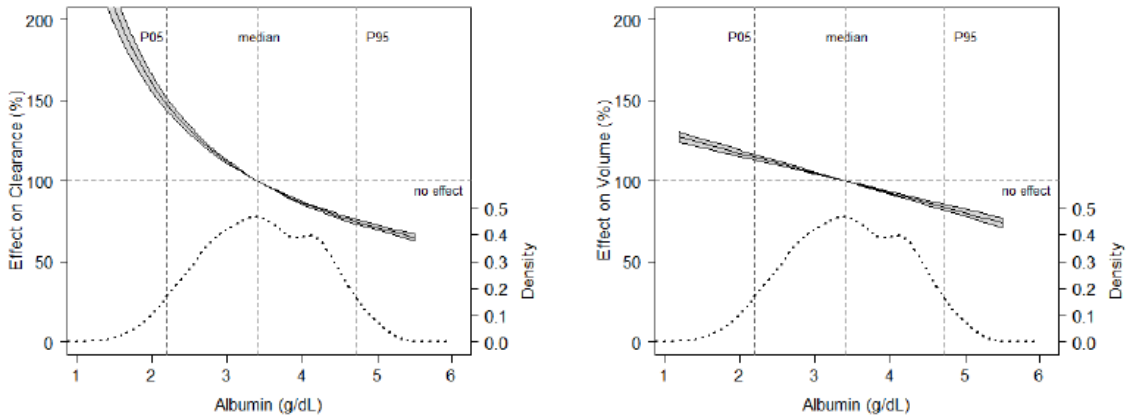



*Source: Applicant's modeling and simulation report, Page 127, Figure 57.*

### *2.1.9.2 Assessment of magnitude of albumin as covariate on CL and Vc*

Albumin effects were included for both CL and Vc. The effect of albumin on CL was described by a power function while the Vc and albumin relationship was described with a linear function. At the P05 (2.2 g/dL) and P95 (4.7 g/dL) of the albumin distribution of the population PK dataset, albumin effects result in a 47.7% increase and 25.2% decrease in CL, respectively, relative to the CL at the median albumin level (3.4 g/dL) (Figure 10). Vc is 14.9% increased and 16.1% decreased at the P05 and P95 of the albumin distribution as compared to the Vc at the median albumin level.

**Figure 10: Distribution of Observed Albumin Levels in the Population PK Dataset and Corresponding Effect on MK-6072 Clearance and Central Volume of Distribution**




*Source: Applicant's modeling and simulation report, Page 128, Figure 58.*

### *2.9.1.3 Assessment of magnitude and adequacy of categorical covariates on clearance and central volume of distribution*

The categorical covariates gender, race (black vs non-black), and Japanese race were included in the final MK-6072 population PK model. The magnitudes of their effects on either CL or Vc of MK-6072 are provided in Table 11.

**Table 11: Magnitude of Categorical Covariate Effects on MK-6072 Clearance and Central Volume of Distribution**

| | Magnitude effect on | |
|---|---|---|
| Covariate | CL | Vc |
| Japanese | 9.5% decreased | 14% decreased |
| Race - Black | 15% increased | - |
| Gender - Male | 22% increased | 24% increased |

Typical subject: White, female, non-Japanese race, body weight 69.7 kg, albumin level 3.4 g/dL

*Source: Applicant's modeling and simulation report, Page 129, Table 26.*

The most pronounced effect was seen for gender, where a typical male subject has 22% higher CL and 24% higher Vc compared to a typical female subject. Black race was associated with a 15% higher CL compared to a typical White subject. Japanese race was associated with a a 9.5% lower CL and 14% lower Vc compared to a typical non-Japanese subject.

*Reviewer's comments: The reviewer verified the Applicant's population PK model for MK-6072. The goodness-of-fit plots indicate that the model reasonably describes the data. The reviewer agrees that no clinically significant impact of body weight, race, gender, and albumin were identified. It is noteworthy that albumin is highly correlated with exposure as well as health condition of patients. The patients with lower albumin level at baseline tend to have lower exposure and sicker health condition. Therefore, albumin becomes a confounding factor for both PK and efficacy/safety endpoint. In addition, the levels of albumin are slightly changing over time, however, the albumin levels at each visit were not provided by the Applicant. The impact of albumin change on exposure is unknown. The reviewer performed independent analysis to assess the accuracy of the labeling language derived from population PK model; the results are consistent with what was proposed by the Applicant as shown in Table 1.The proposed labeling language based on population PK analyses is acceptable from a pharmacometrics perspective.*

## 2.2 Exposure-response analyses

### 2.2.1 Objectives

- To evaluate the E-R for efficacy based on the prevention of recurrence of CDI by MK-6072 as observed in Phase 3.
- To investigate covariates impacting the CDI recurrence after treatment with placebo or active treatment were investigated. The presence of an MK-6072 exposure-response relationship and the type of relationship (linear, non-linear) was then evaluated in a step-wise manner.
- To evaluate the E-R for safety based on the any AE reported in the 4 weeks following infusion and serious AEs reported in the 12 weeks following infusion after treatment with Placebo and MK-6072 in Phase 3.

### 2.2.2 Trial included in the exposure-response analysis

The exposure-response analysis included data from two Phase 3 trials summarized in Table 7. MK-6072 or MK-3415A was administered as single IV infusions of 10 mg/kg over approximately 1 hour.

### 2.2.3 Efficacy endpoint

The primary efficacy endpoint for the Phase 3 trials was the proportion of patients with CDI recurrence at Week 12, defined as the development of a new episode of diarrhea associated with a positive stool test for toxigenic *C. difficile* following clinical cure of the initial CDI episode. A patient was diagnosed with CDI by the presence of diarrhea, as defined by passage of 3 or more loose stools in 24 or fewer hours, and a positive stool test for toxigenic *C. difficile*. CDI was reported at any time during the study. The daily counts of loose stools, as recorded by the patient in the stool count log, were monitored following the infusion through Week 12 in order to identify a new episode of diarrhea. All new episodes of diarrhea were tested for toxigenic *C. difficile* to confirm CDI recurrence.

### 2.2.4 Safety endpoint

The safety endpoint was defined as any adverse event, including clinical adverse experiences (plus infusion-related reactions) and laboratory adverse experiences in the All Patients as Treated (APaT) Population. Non-serious adverse experiences were collected from the time of infusion until Week 4 post-infusion; serious adverse experiences are collected until Week 12. Adverse experiences occurring prior to study therapy administration as a result of protocol-specified procedure or intervention, when reported, were not included in the PK-AE analysis for lack of causality. AEs that occurred in at least 1% of the Phase 3 population were considered for exposure-response analysis.

### 2.2.5 Data analysis plan

Data exploration was conducted using R 2.15.2 (The R Foundation for Statistical Computing, Vienna, Austria).

Potential exposure-response (E-R) relationships for efficacy were explored by plotting the incidence of CDI recurrence in Phase 3 against binned exposure values for MK-6072. Additionally, plots were stratified by covariates (Table 7) to explore the potential impact of covariates of interest.

Exposure-response analyses for safety were conducted for the following endpoints: 1) all adverse events (AEs) reported in the first four weeks following infusion and 2) serious adverse events (SAEs) reported in the 12 weeks following infusion in patients receiving MK-6072, MK-3415A, or placebo in the Phase 3 trials.

### 2.2.6 Exposure-response results

The exposure-CDI recurrence relationship based on $AUC_{0\text{-}inf}$ was best characterized with an Emax model. Of note, the EAUC50 could not be precisely estimated. While EAUC50 could not be precisely estimated, it was assumed to fall between exposures correspond to 0 to < 6 mg/kg. Consequently, EAUC50 was fixed to 100 µg·h/mL, which is well below the exposure range observed upon MK-6072 treatment of 10 mg/kg for subsequent model development. $AUC_{0\text{-}84d}$ and $C_{max}$ were also evaluated as predictors for CDI recurrence, but no improvement in model fit was found compared to $AUC_{0\text{-}inf}$.

The final exposure-response model for CDI recurrence consisted of a placebo effect and an Emax function describing the relationship between CDI recurrence and MK-6072 $AUC_{0\text{-}inf}$. The incidence of CDI recurrence in patients is affected by history of CDI in the past 6 months, albumin, age, and the Charlson Comorbidity Index. The parameter estimates of the final model are presented in Table 12.

The placebo effect and MK-6072 exposure-response Emax were estimated with good precision (%RSE below 16.0%). Precision (%RSE) of the parameters for covariate effects ranged between 15.1% and 42.2%.

**Table 12: Parameter estimates for the final E-R model for CDI recurrence incidence**

| Parameters | Estimate[a] | %RSE[b] | 95% CI[c] |
|---|---|---|---|
| Placebo treatment effect ($b_0$) | -1.15 | 8.37% | -1.34 ; -0.953 |
| Covariates affecting placebo | | | |
| Albumin – continuous | 0.318 | 24.0% | 0.172 ; 0.471 |
| Age – continuous | 0.0103 | 31.7% | 0.00391 ; 0.0166 |
| History of CDI in past 6 months - Yes | 0.707 | 15.1% | 0.513 ; 0.919 |
| Charlson Comorbidity Index – score ≥3 | -0.267 | 42.2% | -0.485 ; -0.0423 |
| MK-6072 | | | |
| Emax | -0.643 | 16.0% | -0.844 ; -0.452 |
| EAUC50 (µg.h/mL) | 100 Fixed | - | - |

*Source: Applicant's modeling and simulation report, Page 145, Table 35.*

The base model development for the MK-6072 exposure – AE relationship consisted of estimation of the placebo effect (baseline probability of AE) and fitting of an exposure-response term. A proportional odds model was utilized to analyze the event as a binominal categorical endpoint (presence of one or more AEs or absence of AE) and applying logistic regression. After estimation of the placebo response, addition of a linear exposure-response relationship for MK-6072 $AUC_{0\text{-}inf}$ did not significantly improve the model fit ($p > 0.05$). MK-6072 $AUC_{0\text{-}84d}$ and $C_{max}$ were also evaluated and similarly resulted in no significant improvement to model fit. Consequently, the exposure-response relationship for AEs in the first 4 Weeks following infusion is characterized by a placebo response term only.

The parameter estimates of the final model are presented in Table 13. Precision (%RSE) of the parameters estimates ranged between 9.1% and 46.1%.

**Table 13: Parameter estimates for the Final E-R model for Adverse Events in the Four Weeks Following Infusion**

| Parameters | Estimate[a] | %RSE[b] | 95% CI[c] |
|---|---|---|---|
| Placebo treatment effect ($b_0$) | 0.239 | 37.5% | 0.049 ; 0.424 |
| Covariates affecting placebo | | | |
| Non-SoC Antibiotics use | 1.03 | 9.1% | 0.845 ; 1.20 |
| Gender – Male | -0.357 | 24.8% | -0.529 ; -0.178 |
| PPI use | -0.362 | 25.2% | -0.552 ; -0.188 |
| Zar score | 0.386 | 33.7% | 0.152 ; 0.649 |
| Hospitalization status | 0.234 | 46.1% | 0.0346 ; 0.422 |

*Source: Applicant's modeling and simulation report, Page 151, Table 42.*

Treatment with MK-6072 appeared to have a similar incidence of SAEs compared to placebo. Exposure-response relationships for incidence of SAEs during 12 weeks following infusion were explored with quantile plots of binned exposure values ($AUC_{0\text{-}inf}$ and $C_{max}$) for MK-6072, and for MK-6072 administered as MK-3415A. In addition, exposure is associated with albumin levels, with low exposure associated with low albumin levels which reflect poorer health and increased likelihood an SAE.

For the evaluation of the exposure-response relationship for SAEs, MK-6072 $AUC_{0\text{-}inf}$ significantly improved model fit ($p < 0.05$), though the directionality of the response indicated a decrease in incidence of SAEs with increasing exposure.

The parameter estimates of the final model are presented in Table 14. Precision (%RSE) of the parameters estimates ranged between 20.7% and 50.1%. While the precision of the age covariates was moderately highly, this covariate was retained in the final model to due to expectation that age impacts incidence of SAEs (i.e. older patients are more likely to have SAEs).

**Table 14: Parameter estimates for the final E-R model for Serious Adverse Events in the Twelve Weeks Following Infusion**

| Parameters | Estimate[a] | %RSE[b] | 95% CI[c] |
|---|---|---|---|
| Intercept ($b_1$) [d] | 5.64 | 33.6% | 2.11 ; 9.56 |
| Covariates affecting placebo | | | |
| Albumin [e] | -0.516 | 29.8% | -0.815 ; -0.209 |
| Non-SoC Antibiotics use [e] | 0.913 | 20.7% | 0.565 ; 1.29 |
| PPI use [e] | -0.649 | 27.1% | -1.01 ; -0.317 |
| Hospitalization status [e] | -0.761 | 32.6% | -1.30 ; -0.286 |
| Charlson's Comorbidity Index [e] | 0.373 | 46.7% | 0.0292 ; 0.719 |
| Age [e] | 0.0114 | 50.1% | 0.0004 ; 0.023 |
| Slope log-linear E-R ($b_2$) [d] | -0.630 | 28.0% | -0.993 ; -0.305 |

*Source: Applicant's modeling and simulation report, Page 157, Table 48.*

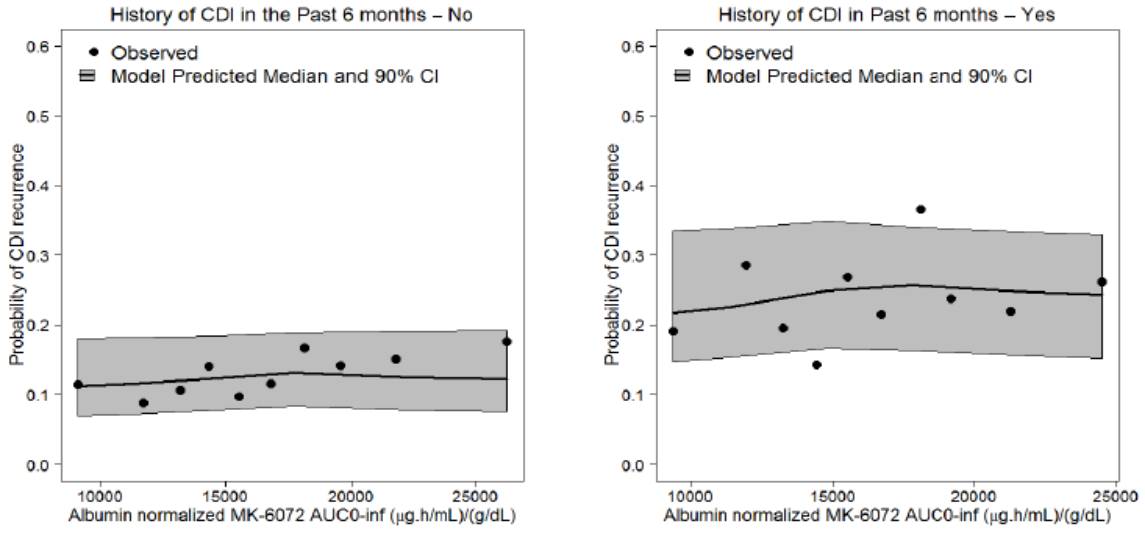
### 2.2.7 Simulations

To visualize the effect of covariates on the relationship between CDI recurrence and exposure over the range of exposure achieved at 10 mg/kg in the Phase 3 trials, simulated and observed CDI recurrence rates were plotted after accounting for the two covariates that had the largest effects on CDI recurrence rate, history of CDI in the past 6 months and albumin level. Albumin level was accounted for by normalizing individual exposures by individual albumin level (i.e. $AUC_{0\text{-}inf}$/albumin), due to the correlation between individual exposures and albumin level. History of CDI in the past 6 months was accounted for by stratification of patient response by this covariate (yes/no).

Figure 11 shows simulated and observed CDI recurrence by albumin-normalized exposure decile, stratified by history of CDI in the past 6 months (yes/no). After adjusting for the effects of these covariates, there were no trends between MK-6072 exposure and observed CDI recurrence rates over the range of exposures achieved with 10 mg/kg in Phase 3. Consistent with this, simulated CDI recurrence rates based on the Emax exposure-response model are flat over the range of exposures associated with 10 mg/kg. This indicates that patient covariates, rather than exposure, are the primary factors influencing CDI recurrence and that the entire range of exposures achieved with 10 mg/kg is associated with a similar, low rate of CDI recurrence.

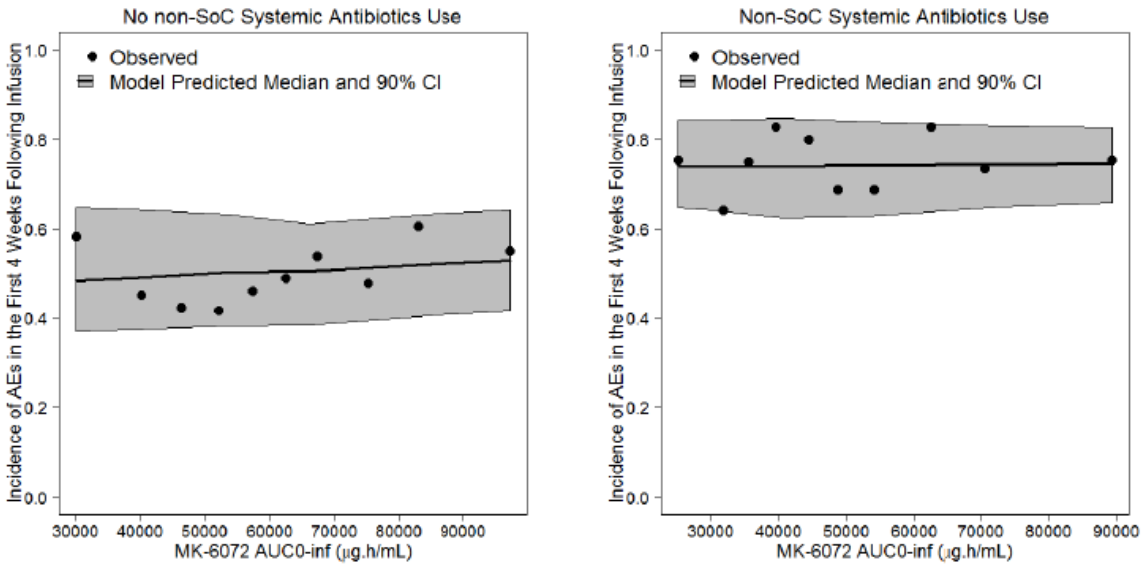
Figure 12 displays simulated and observed incidences of AE occurrence in the 4 weeks following infusion by exposure decile and stratified by non-SoC antibiotic use, the most significant covariate influencing the incidence of AEs. Once covariate effects are accounted for, the incidence of AE occurrence was not dependent on exposure over the range of exposures associated with 10 mg/kg in the Phase 3 trials.

**Figure 11: Maximal Response in CDI Recurrence Rate Achieved Across MK-6072 $AUC_{0\text{-}inf}$ by Decile in Patients Following Administration of a Single IV Dose of 10 mg/kg MK-6072 or 10 mg/kg MK-3415A**




*Source: Applicant's modeling and simulation report, Page 212, Figure 103.*

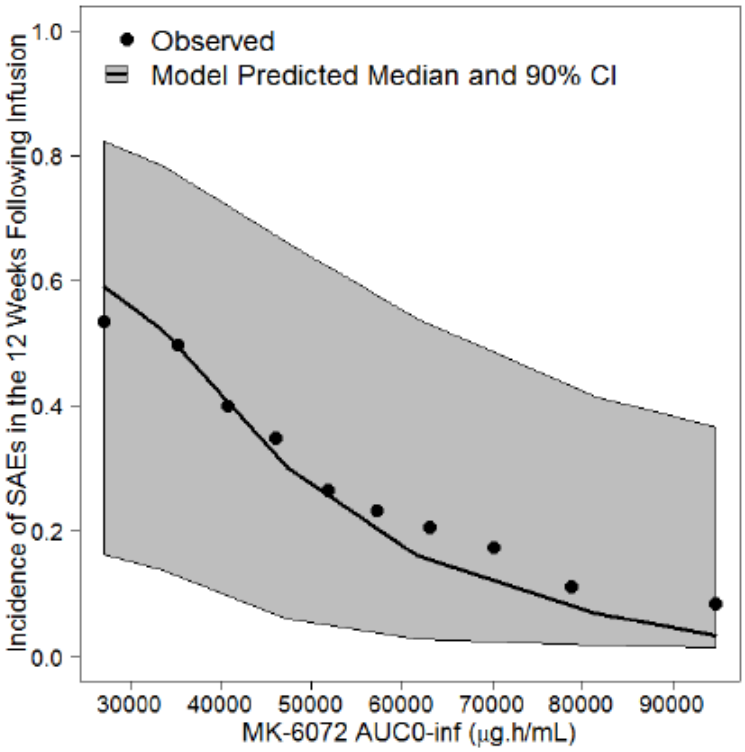
**Figure 12: Relationship Between MK-6072 $AUC_{0\text{-}inf}$ and Incidence of Adverse Events During the 4 Weeks Following Infusion in Patients Following Administration of a Single IV Dose of 10 mg/kg MK-6072 or 10 mg/kg MK-3415A**




*Source: Applicant's modeling and simulation report, Page 215, Figure 105.*

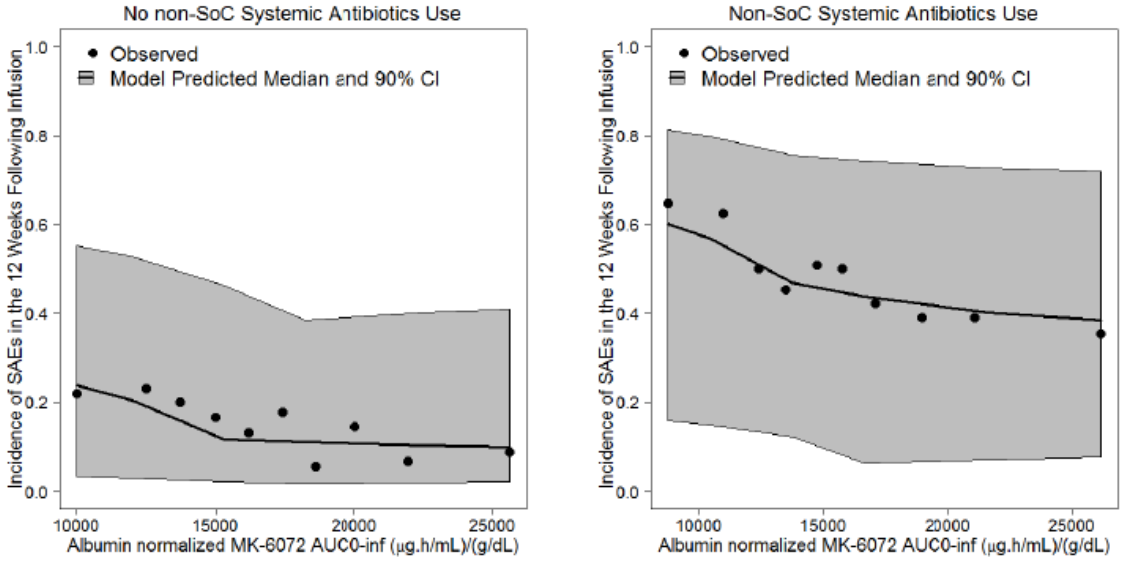
Model-based simulations of the incidence of SAE occurrence were conducted in order to assess the impact of the covariates on SAE occurrence over the range of exposure achieved at 10 mg/kg in the Phase 3 trials. The simulations allowed for plotting the predicted versus observed incidence of SAEs after accounting for the two most influential covariates, namely albumin level and concomitant use of non-SoC antibiotics. Albumin level was accounted for by normalizing individual exposures by individual albumin level (i.e. $AUC_{0\text{-}inf}$/albumin) and concomitant non-SoC antibiotic use was accounted for by stratification of patient response by this covariate (yes/no). Exposure was plotted by decile. After adjusting for the effects of these covariates, the trends between MK-6072 exposure and incidence of SAEs over the range of exposures achieved with 10 mg/kg in Phase 3 (Figure 13) were markedly reduced (Figure 14). In particular, the increase in the incidence of SAEs observed at the lowest exposure decile is attenuated by accounting for these two patient covariates. Thus, these patient covariates account for most of the initially observed exposure-response trend for SAEs.

**Figure 13: Relationship Between the Incidence of Serious Adverse Events in the 12 Weeks Following Infusion Across Increasing MK-6072 Exposure Deciles in Patients Following Administration of a Single IV Dose of 10 mg/kg MK- 6072 or 10 mg/kg MK-3415A**




*Source: Applicant's modeling and simulation report, Page 216, Figure 106.*

**Figure 14: Relationship Between MK-6072 Exposure and Incidence of SAEs During the 12 weeks Following Infusion After Including Patient Covariates in Patients Following Administration of a Single IV Dose of 10 mg/kg MK-6072 or 10 mg/kg MK-3415A**




*Source: Applicant's modeling and simulation report, Page 217, Figure 107.*

*Reviewer's comments: The reviewer performed independently exposure-response analysis for efficacy and safety. Since the patients who experienced clinical initial failure during standard of care was not assessed for the CDI recurrence in the follow-up period, these patients was excluded from our exposure-response analysis for efficacy but not for safety. The reviewer agrees with the conclusion that there were no exposure-response relationships identified for both efficacy and safety. However, the Reviewer thinks it is not reasonable to normalize $AUC_{0\text{-}inf}$ by albumin, as albumin is not only associated with exposure but also an indicator for disease condition. Albumin level should be considered as a confounding factor for PK, PD and safety. Therefore, the Reviewer compared the odds ratios of CDI recurrence from treatment arm to that from placebo arm across the albumin level bins for both efficacy and safety. The odds ratios are flat across the albumin level bins, suggesting that no dose adjustment is necessary based on exposure-response analysis. In addition, as the albumin level is changing over time, the impact of time-varying albumin level on exposure and efficacy/safety is unknown due to inadequate information. If subsequent trials are requested of the Applicant in this population, more comprehensive data collection should be specified as part of the protocol, including tracking of laboratory values over the full course of treatment rather than baseline and week 4 as was collected in these registrational trials.*

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/s/

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