Application Type	Original BLA Application
STN	125690/0
CBER Received Date	July 15, 2019
PDUFA Goal Date	March 14, 2020
Division / Office	DVP /OVRR
Committee Chair	Stephanie L. Polo
Clinical Reviewer	Rebecca Reindel
Project Manager	Richard Daemer, Ph.D.
Priority Review	Yes
Reviewer Name	Charles (Yin Kiu) Cheung
Review Completion Date / Stamped Date	
Concurrence #1	Lei Huang, Concurring Reviewer, Vaccine Evaluation Branch, Division of Biostatistics
Concurrence #2	Tsai-Lien Lin, Chief, Vaccine Evaluation Branch, Division of Biostatistics
Concurrence #3	John Scott, Director, Division of Biostatistics
Applicant	Merck Sharp & Dohme Corp.
Established Name	Ebola Zaire Vaccine, Live
Trade Name	ERVEBO
Pharmacologic Class	Vaccine
Formulation(s), including Adjuvants, etc	VSV-G envelope glycoprotein and substituted with the envelope glycoprotein (GP) of the *Zaire* Ebola virus (Kikwit 1995 strain)
Dosage Form(s) and Route(s) of Administration	1.0 mL dose of ERVEBO intramuscularly.
Dosing Regimen	1.0 mL sterile solution for injection supplied as a single-dose vial
Indication(s) and Intended Population(s)	Active immunization of at-risk individuals 18 years of age and older to protect against Ebola Virus Disease (EVD) caused by *Zaire* Ebola virus.

Table of Contents

GLOSSARY

CMC - Chemistry and Manufacturing Control
(b) (4)
ERVEBO - Ebola *Zaire* Vaccine
EML - European Mobile Laboratory
EUA – Emergency Use Authorization
EVD - Ebola Virus Disease
GP-ELISA - Glycoprotein-based Enzyme-Linked Immunosorbent Assay
LoD – Limit of Detection
PFU - Plaque Forming Units
PRNT - rVSV-ZEBOV-GP Plaque Reduction Neutralization Test
RT-PCR - Reverse transcription polymerase chain reaction
ZEBOV - Zaire Ebola virus

1. Executive Summary

Merck & Co., Inc submitted an original Biologics License Application for ERVEBO, an Ebola *Zaire* Vaccine (rVSVΔG-ZEBOV-GP, live, attenuated; also known as V920) indicated for active immunization of at-risk individuals 18 years of age and older to protect against Ebola Virus Disease (EVD) caused by *Zaire* Ebola virus.

The (b) (4) assay is used for determining the potency of (b) (4) Drug Product during commercial release and stability testing in the manufacturing process. This assay was validated and considered to be acceptable based on a previous review of the validation report under IND 16131/0.116.

Several Phase 2 and 3 clinical trials were conducted to evaluate clinical efficacy and immunogenicity of the vaccine. The Glycoprotein-based Enzyme-Linked Immunosorbent Assay (GP-ELISA) and rVSV-ZEBOV-GP Plaque Reduction Neutralization Test (PRNT) were used for quantifying immunogenicity responses to the vaccine. Validations of GP-ELISA and PRNT assays were previously reviewed under Master File 16537/0.6 and IND 16131/0.127, respectively. These assays were considered to be acceptable for use in clinical studies V920-009, V920-010, V920-011, and V920-012.

In the pivotal clinical study V920-010, Reverse transcription polymerase chain reaction (RT-PCR) assay(s) was(were) used for detection of EVD in human samples. The (b) (4) Ebolavirus RT-PCR Kit (b) (4), when used in conjunction with the PCR instruments listed in the Emergency Use Authorization (EUA), appears to have demonstrated adequate performance in non-clinical settings. In V920-010, the Guinean national surveillance network identified EVD cases using assays that were believed to be the same as or similar to the (b) (4) Ebolavirus RT-PCR Kit (b) (4). After initial identification of cases, the majority of the EVD cases (i.e., aliquots from 79% (93/117) of the EVD index cases and 83% (87/105) confirmed EVD endpoint cases) were confirmed by the European Mobile Laboratory (EML) using the (b) (4) Ebolavirus RT-PCR Kit (b) (4) with either (b) (4), which were not listed as

approved PCR instruments in the EUA for the kit. The retest results performed by EML showed high positive percent agreement with the primary results by the Guinean national surveillance network.

In another experiment, ten clinical samples were tested in eleven labs using combinations of different PCR assay kits, extraction kits, and PCR instruments. Although the number of samples evaluated in the experiment was small, the results from laboratories 2b and 2d, which used the (b) (4) Ebolavirus RT-PCR Kit (b) (4) with the (b) (4) Viral RNA extraction kit and (b) (4) instrument, had reasonable agreement with the results from other laboratories that used different PCR assays, extraction kits, or PCR instruments, except for Sample #5. Of note, laboratories 2b and 2d used the same Ebola PCR assay and PCR instrument as the EML. Nevertheless, no comparison has been performed for the (b) (4) Ebolavirus RT-PCR Kit (b) (4) when used with the EUA-approved PCR instruments and with the (b) (4) PCR instruments (the EML setup). After internal discussion with other members of the review team, we have determined that the results from the "External Quality Assurance Panel" suggest that the EML setup has good agreement with the results from setups with other combinations of assay kits and PCR instruments. Therefore, I believe the determination of EVD cases in V920-010 is sufficiently reliable to support the use of the results of the study as substantial evidence of effectiveness.

2. Regulatory Background

The rVSVΔG-ZEBOV-GP, a live, attenuated vaccine (also known as V920), is genetically engineered to have the gene encoding for the vesicular stomatitis virus glycoprotein G deleted from its RNA and replaced with the gene for *Zaire* Ebola virus glycoprotein. The V920 clinical development program was initiated in 2014, in response to the large Ebola outbreak in West Africa. Eight Phase 1 trials (V920-001 through V920-008) were conducted. A Phase 3 clinical trial V920-010 was used to evaluate the clinical efficacy in Guinea. Phase 2/3 clinical trials V920-009 and V920-011 were conducted to evaluate immunogenicity using validated assays in samples collected from subjects in Liberia and Sierra Leone, respectively. A Phase 3 clinical study V920-012 was performed to evaluate lot consistency and durability of immune response.

2.1 Summary of Pre- and Post-submission Regulatory Activity Related to the Submission

The Investigation New Drug 16131 was initiated in August 2014. The following events were related to my review.
- On February 13, 2017, CBER communicated to Merck Sharp & Dohme Corp that the Plaque Reduction Neutralization Test (PRNT) was suitable for measuring neutralizing antibodies in human serum samples (IND 16131).
- On February 16, 2017, CBER communicated to the United States Army Medical Materiel Development Activity that the GP-ELISA assay validation was acceptable and clinical sample analysis could proceed (Master File Number (b) (4).

3. SOURCES OF DATA AND OTHER INFORMATION CONSIDERED IN THE REVIEW

3.1 Review Strategy

My review was divided into the Chemistry and Manufacturing Control (CMC) and clinical bioassay sections. In the CMC section, I referenced the previous review of the (b) (4) assay for measuring potency, in which the assay was determined to be validated. In the clinical bioassay section, I reviewed the documents related to the assessment of the (b) (4) Ebolavirus RT-PCR Kit, which was used in the efficacy trial V920-010 for determining the case status of Ebola Virus Disease (EVD). In addition, I referenced the previous reviews of the GP-ELISA and PRNT, which were used for measuring immunogenicity responses to the vaccine in the Phase 2 trial V920-009, Phase 2/3 trial V920-011, and Phase 3 trial V920-012.

3.2 Submission Quality and Completeness

Documents related to the (b) (4) Ebolavirus RT-PCR Kit were submitted. A document containing the "Instruction for Use" was submitted. This document provided information about the performance of the assay, and the description was generally clear. Additional evaluations performed on the assay were presented in the documents entitled "Clinical Information Amendment" and "External Quality Assurance Panel for Ebola RT-PCR Field Diagnostics - GUINEA" submitted to BLA 125690/0.18.

3.3 BLA/IND Documents That Serve as the Basis for the Statistical Review

The documents under review were:
1. BLA 125690/0.18 dated 5/22/2019
    - Module 1.11.3 *Clinical IR Response 27 March 2019*
    - Module 1.11.3 *1 PCR Response EMLab_EQA_Report-for-all-labs*
    - Module 1.11.3 *2 PCR Response* (b) (4) *-Zaire-Ebolavirus-RT-PCR-Kit-* (b) (4) *_Precision*
    - Module 1.11.3 *3 PCR Response Analytical Plan*
    - Module 1.11.3 *4 PCR Response EML-M-034 Setup* (b) (4) *Ebolavirus RT-PCR*
    - Module 1.11.3 *5 PCR Response EML-M-042-A1-* (b) (4)
    - Module 1.11.3 *6 PCR Response EML-M-043 Validation of real time RT-PCR results*
    - Module 1.11.3 *7 PCR Response Commissioner signature EUA Letter_med_11-26-14*
    - Module 1.11.3 *8 PCR Response EML-M-034-B* (b) (4) *Ebolavirus RT-PCR kit* (b) (4)

4. CHEMISTRY, MANUFACTURING, AND CONTROLS

4.1 Chemistry, Manufacturing, and Controls

(b) (4) **assay:**

The (b) (4) assay is used to measure the potency of the (b) (4) Drug product. This assay is used for commercial release and stability testing and was developed at Merck and validated at (b) (4) . The validation report entitled "Technical Communication for the Validation of V920 (b) (4) Assay at (b) (4) , WP75A, Oct-2016" was previously reviewed by a statistical reviewer under IND 16131/0.116. The assay characteristics evaluated against pre-defined acceptance criteria during validation were accuracy, repeatability, intermediate precision, linearity, and range. The statistical reviewer concluded that "the data appear to support that the assay has been validated."

5. CLINICAL BIOASSAYS

The (b) (4) Ebolavirus RT-PCR Kit (b) (4), GP-ELISA, and PRNT were used to assess the primary efficacy and immunogenicity endpoints in the pivotal studies, and the results were presented in the package insert of ERVEBO.

5.1 RT-PCR assay for the detection of Ebola virus disease (EVD)

The (b) (4) Ebolavirus RT-PCR Kit was authorized under Emergency Use Authorizations (EUA) by FDA on (b) (4) . The test was intended for qualitative detection of RNA from Ebola virus (such as (b) (4)

on specified instruments in (b) (4) plasma from individuals with signs and symptoms of Ebola virus infection in conjunction with clinical and epidemiological risk factors. The (b) (4) Ebolavirus RT-PCR Kit (b) (4) is intended for use only under the Emergency Use Authorization (EUA) by laboratories certified under the Clinical Laboratory Improvement Amendments of 1988, 42 U.S.C. §263a, to perform high complexity tests, or by similarly qualified non-U.S. laboratories, and is limited to clinical laboratory personnel specifically instructed and trained in the techniques of real-time PCR and *in vitro* diagnostic procedures. Information about this test kit was previously submitted to IND 16131/0.118 and was resubmitted to BLA 125690/0.18.

Method description:

The (b) (4) is used to extract RNA from a sample. Then, the extracted RNA is tested using the (b) (4) Ebolavirus RT-PCR Kit. This test consists of (b) (4) steps: (b) (4)

. To perform PCR amplification, (b) (4)

(b) (4)   can be used. The
PCR targets the (b)(4) gene of the Ebola virus.

Analytical performance:
Analytical Sensitivity (Determination of the Limit of Detection (LoD)):
Experiment:

- The Limit of Detection (LoD) of (b) (4)   was
  confirmed by (b) (4)

Results:

- (b) (4) (95%; 95% CI: 75.1% - 100%), (b) (4) (100%; 95%CI: 83.2%-100%), and
  (b) (4) (95%; 95%CI: 75.1% - 100%) replicates were detected positive when the
  RT-PCR kit was used with the (b) (4)
                                                  , and (b) (4)   , respectively.

Reviewer's comment:

- *Across the (b)(4) PCR instruments used with the (b) (4) Ebolavirus RT-PCR Kit
  (b) (4), the point estimate of sensitivity was at least 95% (95% lower confidence limit
  of (b) (4), evaluated at the LoD of (b) (4) using the (b) (4)
                           strain. Assuming data from the (b) (4) systems can be
  pooled, (b) (4) was detected in (b) (4) out of (b) (4) samples, with a detection rate of
  (b) (4) (95% CI: (b) (4)   However, the evaluation only assessed a (b) (4)
  ebolavirus strain.*

Analytical Specificity:
Specificity was assessed in terms of different *ebolavirus* strains (reactivity), different
viruses (cross-reactivity), negative samples, and interfering substances.

A. Reactivity:
The (b) (4) Ebolavirus RT-PCR Kit with (b) (4)
was tested against different Ebolaviruses (i.e., (b) (4)

                                                          presumably (b) (4) per
strain.
Results: All samples were tested positive.

B. Cross-reactivity
Genomic RNA/DNA of a diverse set of pathogens were analyzed with the (b) (4)
Ebolavirus RT-PCR Kit on (b) (4)   . The list of
pathogens is:

- (b) (4)

(b) (4)

Results: No cross-reactivity was observed (i.e., all results for Ebolavirus were negative).

C. Negative samples
A total of (b) (4) negative plasma samples from individual donors were tested by the kit on the (b) (4) system, and (b) (4) respectively.

Results: All results for Ebolavirus were negative.

D. Interfering substances
Each interfering substance (i.e. (b) (4)

) were spiked into simulated (b) (4) plasma samples containing no Ebolavirus, Ebolavirus in Positive Control Target EBOLA (approximately (b) (4) LoD), and Ebolavirus in Positive Control Target EBOLA (b) (4) diluted (approximately (b) (4) LoD), separately. Each sample was tested in (b) (4) replicates. The samples were tested by the kit with the (b) (4) system.

Results:
- All negative samples were tested to be negative.
- All ~(b) (4) LoD samples were tested to be positive.
- All ~(b) (4) LoD samples were between (b) (4) in mean C_p value, where C_p is the cycle at which fluorescence achieves a defined threshold for case detection.
  - Within (b) (4) difference, relative to the mean C_p value of the reference sample

Reviewer's comment:
- *The assessment of specificity in terms of different ebolavirus strains (reactivity), different viruses (cross-reactivity), negative samples, and interfering substances provides evidence of acceptable specificity.*

Clinical performance evaluation: Sensitivity and Specificity
Experiment:
Mock clinical samples were used to assess the clinical sensitivity and specificity.
First, viral RNA (presumably for the (b) (4)

(b) (4)

(b) (4)

Ebolavirus RT-PCR Kit on the (b) (4) system and (b) (4) , respectively. No acceptance criteria were pre-specified.

Results:

- For tests performed using the (b) (4) and (b) (4) system, all (b) (4) positive samples (b) (4) LOD, (b) (4) LOD, and (b) (4) LOD) were tested positive. All (b) (4) negative samples were tested negative.
- For testing performed using the (b) (4) out of (b) (4) LOD samples, all (b) (4) LOD and all (b) (4) LOD samples were tested positive. All (b) (4) negative samples were tested negative.
- Positive Percent agreement (PPA) and Negative Percent Agreement (NPA) were summarized in Table 1.

(b) (4)

Reviewer's comments:

- *In this experiment, viral RNAs were spiked into mock samples. Because the true statuses of the samples were known, what the applicant refers to as "Positive Percent Agreement" and "Negative Percent Agreement" are more commonly referred to as "sensitivity" and "specificity."*
- *The results appear to demonstrate that the kit has reasonable clinical sensitivity and specificity for samples with concentration* (b) (4) *LoD and above for the mock* (b) (4) *strain.*

Comparison of the (b) (4) Ebolavirus RT-PCR Kit (b) (4) to published tests:
Experiment:

- For each of the (b) (4) virus (b) (4)

These samples were tested using (b) (4) different methods (b) (4) . Each sample was tested (b) (4) times for the (b) (4) Ebolavirus RT-PCR Kit (b) (4) and (b) (4) , and (b) (4) for (b) (4) .

Results:

- (b) (4)

Reviewer's comments:

- *The sample size used for each evaluation was small. However, for each strain, all* (b)(4) *test kits appear to make positive determinations consistently when the stock concentration is high enough.* (b) (4) *Ebolavirus RT-PCR Kit* (b)(4) *and* (b) (4) *appear to have a lower LoD compared to the* (b) (4) *.*
- *The* (b) (4) *Ebolavirus RT-PCR Kit did not make consistent positive determinations for the* (b) (4) *sample that was diluted to a final concentration of* (b) (4) *(i.e.,* (b) (4) *of the tests were positive). Because* (b) (4) *was close to the established LoD of* (b) (4) *, this data may suggest varying sensitivity of the* (b) (4) *near the LoD. Nevertheless, the sample size was too small to draw firm conclusions.*

External experiments:
External quality assurance panel

A document entitled "External Quality Assurance Panel for Ebola RT-PCR Field Diagnostics – GUINEA" was submitted. The document contains a table that presented the test results of (b)(4) samples, tested by (b)(4) distinct lab IDs. (b) (4) laboratories (b) (4) used the (b) (4) Ebolavirus RT-PCR Kit, (b) (4) laboratories used the (b) (4) RT-PCR kit, (4) laboratory used the (b) (4) assay kit, (4) laboratory used the (b) (4) rRT-PCR kit, (4) laboratory used the (b) (4) kit, and (4) laboratory used the (b) (4) kit.

Reviewer's comment:

- *As of today, the* (b) (4) *Ebolavirus RT-PCR,* (b) (4) *rRT-PCR, and* (b) (4) *assay kits are approved for use by the FDA under the EUA.*
- *Laboratories* (b) (4) *used the* (b) (4) *Ebola RT-PCR kit with* (b) (4) *real-time PCR and* (b) (4) *platforms, respectively, instead of one of the PCR instruments that were authorized by the EUA for this kit (Table 2).*

These (b) (4) *labs used the same combination of extraction kit, RT-PCR kit and PCR instrument as the EML.*

Results:
- The results of testing across laboratories were presented in Table 2.
    - (b)(4) samples were tested negative by all laboratories.
    - (b)(4) samples were tested positive by at least (b) (4) laboratories.
        - (b) (4) of the (b) (4) laboratories that used the (b) (4) Ebola RT-PCR Kit obtained a negative result on sample (b) (4). However, the sample was tested positive by (b) (4) other laboratories. The (b)(4) laboratories that had negative results all used the (b) (4) for the PCR amplification step.
        - For all (b)(4) other positive samples, the (b) (4) laboratories that used the (b) (4) Ebola RT-PCR Kit obtained positive results.

Reviewer's comment:

- *Among the laboratories that tested the samples using the* (b) (4) *Ebola RT-PCR kit, laboratory* (b) (4) *agreed with the determination from a majority of laboratories in all* (b) (4) *samples* (b) (4), *while laboratory* (b) (4) *agreed with the determination from a majority of laboratories in* (b) (4) *out of* (b) (4) *samples* (b) (4)) (b) (4) *negative percent agreement and* (b) (4) *positive percent agreement with the majority]. Of note, Labs* (b) (4) *used the same Ebola PCR assay and PCR instrument as the EML. This assessment provided additional evidence that the EML assay* (b) (4) *Ebola RT-PCR kit when used with* (b) (4) *or* (b) (4) *instrument) generates similar test results with several other commercial kits.*

Retests of Clinical Samples by EML

Clinical study V920-010 (Ring vaccination study) was a Phase 3, open-label, cluster-randomized trial using ring vaccination to evaluate efficacy and safety of ERVEBO in Guinea. The primary objective of this pivotal clinical study V920-010 was to assess vaccine efficacy against laboratory-confirmed Ebola virus disease (EVD) by comparing immediate versus delayed ring vaccination. The primary outcome was confirmed EVD, which was defined as any probable or suspected case laboratory confirmed as positive for EVD, or any deceased individual with probable EVD, from which a post-mortem sample taken within 48 hours after death was laboratory confirmed as positive for EVD. Laboratory confirmed cases must be tested positive for the virus by detection of virus RNA by RT-PCR.

Laboratory testing was performed by national and international laboratories in the Guinean national surveillance network. Testing was performed with blinding; laboratories ignored the vaccination status or allocation arm of the suspected case. The applicant stated that the same or similar assays were used by the Guinean national surveillance network and the European Mobile labs (EML): (b) (4) Ebolavirus RT-PCR Kit (b) (4)

Confirmation of positive cases:

The EVD test results generated by the Guinean national surveillance network laboratories represented the primary laboratory confirmation. If available, aliquots were retested at EML using the (b) (4) Ebolavirus RT-PCR kit (b) (4). The EML used (b) (4) or (b) (4) real-time PCR platform instead of one of the PCR platforms specified under FDA's letter of authorization for emergency use of the (b) (4) Ebolavirus RT-PCR Kit.

Overall, there were 117 EVD index cases and 105 confirmed EVD cases (34 cases with onset of 10 or more days after randomization and 71 cases with onset of less than 10 days after randomization) recorded in the study. The EML obtained aliquots from 79% (93/117) of EVD index cases; 88% (30/34) of confirmed EVD outcome cases with onset of 10 or more days after randomization and 80% (57/71) of all confirmed EVD outcome

cases with onset of less than 10 days after randomization. Five cases of EVD initially considered as index cases for clusters were negative by confirmatory testing at the EML. These corresponding clusters were excluded from the analysis in the clinical study. No endpoint cases were tested negative in the EML retesting.

Reviewer's comment:
- *The applicant was not able to confirm whether the* (b) (4) *Ebolavirus RT-PCR Kit was used by the Guinean national surveillance network. However, it was observed that 93 EVD cases retested positive by the EML, which used the* (b) (4) *Ebolavirus RT-PCR Kit, and only 5 EVD cases initially considered as index cases for clusters retested negative by the EML. Thus, the test results at the Guinean laboratories and EML appear to have reasonably good positive percent agreement (88/93=95% for index cases, and 30/30=100% for endpoint cases).*

5.2 Immunogenicity assays

GP-ELISA:
The GP-ELISA was used to quantify the total IgG antibodies against Zaire glycoprotein. The assay uses purified Zaire Ebola virus recombinant glycoprotein (ZEBOV-rGP) as the coating antigen and an enzyme-conjugated anti-human IgG secondary antibody as the reporter or signal system. The GP-ELISA was previously reviewed by a statistical reviewer under Master File (MF) (b) (4). The GP-ELISA assay was validated by (b) (4) in the documents #AVAL.119.00116 entitled "Validation of Zaire ebolavirus (ZEBOV) IgG ELISA" (MF (b) (4) and #AVAL.119.00156 entitled "Specificity of Elevated Background Sera for ZEBOV IgG ELISA" (MF (b) (4) . CBER sent the following comment to the Department of the Army on February 16, 2017.
- "We have completed our review of MF (b) (4) Amendment 10, submitted on February 7, 2017, including your response to the CBER information request dated November 18, 2016. We agree that the ZEBOV IgG ELISA is adequate for its intended use, and testing of human samples at (b) (4) may proceed. However, the clinical immunogenicity endpoints and the clinical study success criteria will be discussed with each vaccine manufacturer."

Plaque Reduction Neutralization Test (PRNT):
To measure and quantify neutralizing antibodies against V920, Merck and New Link Genetics in collaboration with (b) (4) have developed and validated a PRNT which uses V920 as the inoculating virus. Determination of the neutralizing titer is based upon the percent reduction in viral plaques in the presence of serum compared to that of the virus control without serum. The results are reported as PRNT[(b) (4)], which is the reciprocal of serum dilution that results in a (b) (4) reduction in the number of plaques.

The PRNT was previously reviewed by a statistical reviewer under IND 16131/0.127. In that amendment, the sponsor submitted reports of the validation and robustness evaluation for the TSOP.119.00772 "rVSV-ZEBOV-GP Plaque Reduction Neutralization Test (PRNT)" which was developed to quantify ZEBOV neutralizing antibodies in human serum from individuals who have received the V920 vaccine. These documents

were submitted to the current BLA 125690/0.2. The statistical reviewer concluded that "the assay validation analyses and conclusions appear acceptable". In addition, on February 13, 2017, CBER communicated to Merck Sharp & Dohme Corp the following statement:

- We have completed our review of IND 16131 Amendment 127, submitted on January 12, 2017, which included the Validation Report, AVAL.119.00122, "Validation of rVSVΔG-ZEBOV-GP (Zaire ebolavirus) Plaque Reduction Neutralization Test (PRNT)". We find the validation report and conclusions to be acceptable, and agree that the assay is suitable for measuring neutralizing antibodies in human serum samples."

6. CONCLUSIONS

6.1 Statistical Issues and Collective Evidence

- The (b) (4) assay for measuring potency was previously reviewed, and the assay was considered by the statistical reviewer to be acceptable for use.
- For detection of EVD cases, the test results from EML, which used the (b) (4) Ebolavirus RT-PCR Kit with (b) (4) or (b) (4) real-time PCR platforms, had good positive percent agreement with the test results from the Guinean national surveillance network.
  - ➤ Assessment of analytical and clinical performance appears to suggest that the (b) (4) Ebolavirus RT-PCR Kit has adequate performance for detecting EVD cases, when used with (b) (4) real-time system.
  - ➤ In another experiment, (b) (4) clinical samples were tested in (b) (4) labs using combinations of different PCR assay kits, extraction kits, and PCR instruments. Although the number of samples evaluated in the experiment was small, the results from laboratories (b) (4) , which used the (b) (4) Ebolavirus RT-PCR Kit (b) (4) with the (b) (4) Viral RNA extraction kit and (b) (4) PCR instrument, had reasonable agreement with the results from other laboratories that used different PCR assays, extraction kits, or PCR instruments, except for Sample (b) (4). Of note, laboratories (b) (4) used the same Ebola PCR assay and PCR instrument as the EML. Thus, this assessment provided additional evidence that the EML's assay would produce similar test results as several other commercial kits. Nevertheless, no comparison has been performed for the (b) (4) Ebolavirus RT-PCR Kit (b) (4) when used with the EUA-approved PCR instruments and with the (b) (4) PCR instruments (the EML setup). Therefore, it is unclear whether the EML PCR assay performance can be expected to be similar.
  - ➤ The GP-ELISA and PRNT for measuring immunogenicity were previously reviewed. These assays were considered acceptable for use.

6.2 Conclusions and Recommendations

In conclusion, the (b) (4) assay appears to be adequately validated, so the assay is fit for use in determining the potency of the (b) (4) Drug Product for commercial release and stability testing.

The GP-ELISA and PRNT appear to have adequate performance for use in clinical studies V920-009, V920-010, V920-011, and V920-012.

After internal discussion with other members of the review team, we have determined that the results from the "External Quality Assurance Panel" suggest that the EML setup has good agreement with the results from setups with other combinations of assay kits and PCR instruments. Therefore, I believe the determination of EVD cases in V920-010 is sufficiently reliable to support the use of the results of the study as substantial evidence of effectiveness.