



Food and Drug Administration
Center for Biologics Evaluation and Research
10903 New Hampshire Avenue
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REVIEW MEMORANDUM

STN: 125588

Date: June 1, 2017

Reviewer: Erica Silberstein, Ph.D., CBER/OBRR/DETDD/LEP

To: Iliana Valencia, CBER/OBRR/IO

Through: Alain Debrabant, Ph.D., CBER/OBRR/DETDD/LEP
Sanjai Kumar, Ph.D., CBER/OBRR/DETDD/LEP

Sponsor: Oxford Immunotec, Inc.
700 Nickerson Road, Suite 200
Marlborough, MA 01752

Product: *Babesia microti* Nucleic Acid Test

Type of the Submission: Biological License Application (BLA)

Disciplines Reviewed: Pre-clinical studies
Chemistry, Manufacturing and Controls

Recommendation: I recommend issuing a Complete Response Letter including my letter ready comment (page 11).

I. Description of the test:

The *Babesia microti* Nucleic Acid Test (NAT) is a blood screening (b) (4) test for the detection of specific DNA to *Babesia microti* in whole blood samples collected using EDTA collection tubes.

The test is based on (b) (4). Sample's DNA is purified using the (b) (4) automated nucleic acid purification instrument and then amplified using the (b) (4). The assay employs the following controls:

- Internal endogenous control (Human 18S rRNA)

- *B. microti*-specific controls (high and low positive controls), consisting of *B. microti* infected (b) (4) whole blood diluted in negative human whole blood.
- Negative control, that consists of *B. microti* negative human whole blood.
- No-template control.

The assay allows the analysis of blood specimens collected in EDTA collection tubes and can be used as a stand-alone blood screening application for testing of blood donors and blood donations for evidence of *B. microti* infection.

Since there is currently no FDA licensed test for the diagnosis of babesiosis, this BLA qualified to be as Priority Review.

II. Review comments

A) Oxford Immunotec response to September 29 2015 FDA Complete Response Letter

After extensive review of this application, FDA sent a Complete Response Letter to the sponsor dated September 29, 2015.

Responses to FDA's CR letter were received on December 14, 2016 (Amendment 13). My review focused on the evaluation of the sponsor's responses to Questions 8-16 (Pre-clinical studies) and Questions 18-26 (Chemistry, Manufacturing and Controls), and my comments to the responses that I did not find acceptable are provided below.

- Pre-clinical Studies:

FDA question #8

*The analytical specificity/cross reactivity study has been conducted using seven bacteria species and one yeast species. Please expand the cross-reactivity studies to include the following pathogens (*Plasmodium sp.*, *Leishmania sp.*, *Trypanosoma cruzi*, and *Borrelia burgdorferi*) as agreed upon in the IND study protocols (IND # 14532).*

Oxford Immunotec response to question #8

The sponsor explained that the studies were expanded to include *Plasmodium sp.*, *Trypanosoma cruzi*, and *Borrelia burgdorferi*. The results indicated that none of the tested organisms showed cross-reactivity in the *Babesia microti* NAT.

No testing of *Leishmania sp.* blood samples was performed.

The sponsor did not provide the information requested by FDA. Oxford Immunotec should make all possible efforts to include *Leishmania sp.* DNA samples to evaluate the specificity of the test.

FDA Question #12

In SOP LAB-MFG-10, it is indicated that (b) (4) Babesia parasites/ml were used as low positive control in the NAT assay. The 95% detection limit of NAT is (b) (4) Babesia parasites/ml. The low positive is almost (b) (4) times the limit of detection (LOD). The use of low positive control that is (b) (4) LOD fails the purpose of the low positive control (i.e., to determine if the assay is working as per specifications). FDA recommends that you add an appropriate low positive control to your panel that is close to LOD (b) (4) and submit the results to FDA for review.

Oxford Immunotec response to question #12

The sponsor prepared a new low positive control containing (b) (4) parasites/ml that was tested in (b) (4) extraction runs failure rate of (b) (4)

The results of those studies are not provided.

Oxford Immunotec should provide those study results since the low positive control is a critical component of the assay.

FDA sent a new IR on February 17 2017 and requested this information again in Question #5. My comments on the sponsor's response are included on page 5 of this review memorandum.

- Chemistry, Manufacturing and Controls

FDA Question #25

Please provide the physicochemical acceptance criteria for the purchased oligonucleotides and documentation that the acceptance criteria were met for purity, sequence, and concentration.

Oxford Immunotec response to question #25

The sponsor indicated that acceptance criteria for purity, sequence, and concentration for the purchased primers and probes are shown in Tables 25.1-4.

The requirement for purity is described as (b) (4) and there is no specification indicated for the *Babesia microti* and Human 18s primers.

The sponsor did not provide the information requested by FDA. The response is not acceptable.

FDA sent a new IR on February 17 2017 and requested this information again in Question #13. My comments on the sponsor's response are included on page 10 of this review memorandum.

B) Oxford Immunotec response to February 17 2017 FDA Information Request

After review of Amendment 13, the committee issued a new information request which was sent on February 17, 2017. The sponsor submitted responses to questions 1-8 on March 1, 2017, and to questions 9-23 on March 23, 2017. I reviewed sponsor's responses to Questions 3-12 (Pre-clinical studies) and Question 13 (Chemistry, Manufacturing and Controls),

- Pre-clinical Studies:

FDA Question #3

In your NAT Amendment response received December 14, 2016 to FDA question #8 (CR dated September 29, 2015) regarding the precision study, you have submitted the new precision study. Please clarify the following regarding this study:

- a. The document "Protocol for Estimating Precision of the Babesia microti (DOC-PRO26)" is not dated. Please clarify when the protocol was implemented and when the study was conducted.*
- b. Please clarify if the "component lots" used for this study were released as "finished device lots" before they were used in this study. Please provide the finished device lot numbers and the release dates.*

Oxford Immunotec response to question #3

- a. The sponsor indicated that the protocol was approved on 10/12/16 and implemented on 10/13/16 and that the study was conducted between October and November, 2016.
- b. Oxford Immunotec explained that the component lots used for this study were not released as finished device lots. All components used were released independently from one another in accordance with their respective release protocols. Table 2 (Page 3) shows the component lots used in the NAT Precision Study.

The sponsor provided the information requested by FDA. The responses are acceptable.

FDA Question #4

In your NAT Amendment response received December 14, 2016 to FDA question #9 (CR dated September 29, 2015) regarding cross reactivity studies and question # 10 (CR dated September 29, 2015) regarding interference studies, you have submitted the new cross-reactivity and interference studies.

Please clarify the following regarding these studies:

- a. When the protocols were approved (implemented) and when these studies were conducted?*

- b. *Please clarify if the “component lots” used for these studies were released as “finished device lots” before they were used in these studies.*

Oxford Immunotec response to question #4

a. The sponsor provided the following information:

1. DOC-PRO-28 is the protocol that describes the cross reactivity studies referenced in question #9 which was approved on 6/15/15 and implemented on 6/16/15. The new cross reactivity studies were performed in September, 2016.
2. DOC-PRO-31 is the protocol for endogenous interference studies referenced in question #10 which was approved on 10/21/16 and implemented on 10/27/16. The new interference studies were performed in October, 2016.

Document details for both protocols are provided in Attachments 4.1 and 4.2.

- b. The component lots used for this study were not released as finished device lots. All components used were released independently from one another in accordance with their respective release protocols.

The sponsor provided the information requested by FDA. The responses are acceptable.

FDA Question #5

In your NAT Amendment response received December 14, 2016 to FDA question #12 (CR dated September 29, 2015) regarding incorporating a low positive control of (b) (4) LOD, you have stated that a low positive of (b) (4) LOD was introduced effective 03/26/16. Please provide the design, verification and validation documents that describe how this change was incorporated in the finished device.

Oxford Immunotec response to question #5

The study conducted on November 2, 2015 is provided in Attachment 5.1 (DOC-RPT-88, Report on the Adjustment of the *Babesia microti* NAT Low Positive Control).

The sponsor provided the information requested by FDA. The response is acceptable.

FDA Question #6

In your NAT Amendment response received December 14, 2016 to FDA question #15 (CR dated September 29, 2015), you have provided the information on different lots used in pre-clinical studies. You have submitted studies that were conducted in response to the complete response letter.

- a. Please clarify which studies were carried out with finished device lots (following the definition of finished device lot). Please provide the lot numbers of finished device lots used in this study.*
- b. Please clarify when the first finished device was released/manufactured. Please provide the list of finished device lots that have been released/manufactured since defining the finished device.*

Oxford Immunotec response to question #6

- a. There have been (b) (4) NAT Finished Device Lots released: (b) (4). Table 3, Page 5, shows a complete list of the component lots numbers. A validation report is included in Attachment 7.4, DOC-RPT-58: "Validation Report of the Assembly and Final Release of a *B. microti* NAT System Finished Device Lot".
- b. The sponsor indicated that the first finished device lot is (b) (4) released on February 10, 2017. Table 3, Page 5, shows all finished device lot assembly, release and expiration dates.

The sponsor provided the information requested by FDA. The responses are acceptable.

FDA Question #7

In your NAT Amendment response received December 14, 2016 to FDA question #16 (CR dated September 29, 2015) on process validation, you have provided the validation plan along with process validation report summarizing the results. Please provide the following information:

- a. Please provide a list of critical lab SOPs/Protocols (including version number) that have been locked (no major modification) following process validation.*
- b. Please clarify since when (specific date) NAT manufacturing and testing are being conducted following implementation of process validation (i.e., locked down, with no major changes).*
- c. Please clarify when the document LAB-VAL-19 was implemented.*
- d. Please provide the following documents: LAB-RPT-54, 55, 58 and 62; LAB-VAL-20 and 24. Please ensure that these documents are signed and dated.*

Oxford Immunotec response to question #7

- a. The sponsor provided the requested list in Table 4, List of Critical Lab SOPs/Protocols, Page 6, column “Locked Manufacturing Procedure Revision”, shows the SOPs/Protocols that have been locked.
- b. The information is provided in Table 4, List of Critical Lab SOPs/Protocols, Page 6.
- c. Oxford Immunotec explained that LAB-VAL-19 was approved and implemented on 12/8/16.
- d. Attachments 7.2-7.7 contain the requested reports. Activation date and who approved document revision are provided at the end of each attachment.

The sponsor provided the information requested by FDA. The responses are acceptable.

FDA Question #8

In your NAT Amendment response received December 14, 2016 to FDA question #17 (CR dated September 29, 2015) on the finished device, you have provided an SOP (LABACQ-MOL-102) “Babesia microti NAT Finished Device Lot Final Release Testing”. Please clarify the following regarding this SOP:

- a. *Please clarify when this SOP was approved and implemented.*
- b. *On Page 3 of 15 (P385) of this document the column 1 of the table has “finished device lot components” and column 4 refers to “previously approved.” While the first seven components (that includes primers, probes and master mix) have a selection option of “Yes/No”; the last three rows (that include negative, high and low positive controls) the selection option is “n/a.” Please clarify why the last three rows are different.*
- c. *On Page 7 of 15 (P393), under procedure assay controls it is stated, “Babesia positive (High: (b) (4) and Low: (b) (4) and negative (b) (4) controls will be extracted both test and reference extraction kits, as detailed in the finished device lots. Babesia high positive control eluate (b) (4) will be amplified and detected for inclusion as a component in the finished device lot.” Throughout the submission Babesia high positive control has part (b) (4) . Please clarify which high positive control with part (b) (4) is referred to here. Is high positive control (b) (4) a panel member for finished device release?*

Oxford Immunotec response to question #8

- a. The sponsor indicated that LAB-AQC-MOL-102 has not been approved or implemented and remains in draft. All affected documents will be implemented together prior to the next scheduled manufacturing.

- b. Oxford Immunotec stated that the determination of previous approval of a component is in place to ensure appropriate status labeling, as either 'Approved' or 'Rejected'. This procedure is applicable for incoming primers, probes, master mix and extraction kits, as final release testing serves as the components initial functional test and approval. Upon first approval of final release testing in a finished device lot, components will be labeled as 'Approved'.

NAT high positive, low positive and negative controls are initially tested following SOPs LABMFG-10, LAB-MFG-12 and LAB-MFG-36, in which controls are labeled as 'Approved' or 'Rejected'. Controls may then be included in a finished device lot combination for final release testing. Therefore, a status labeling of these controls in LAB-AQC-MOL-102 is not applicable.

- c. The sponsor explained that (b) (4) is an in-process material utilized in the manufacture of (b) (4) is also utilized in finished device lot release testing and as whole blood control for extraction, ensuring a high positive sample is effectively extracted by incoming extraction kits.

The responses are acceptable.

FDA Question #9

In your NAT Amendment response received December 14, 2016 to FDA question 8a (IR dated November 11, 2016) for samples (b) (6) (that have only (b) (4) PCR repeat testing result instead of (b) (4) results), you have stated that “The initial Babesia Ct value was noted on the test run as a non-exponential curve. Per testing SOP, LAB-MOL-BPCR-10, Section 8.2, a non-exponential curve is considered negative and does not require repeat testing.” However in the document MSTDONOR_PROSPECTIVE (Attachment-2.2_ MSTDONOR_BCR-NAT-ATT-6) under PCR Repeat testing column the result is reported as “Undet/26.8 and Undet/23.4”.

- a. *Please clarify if the final result designating the samples as negative was based on initial testing and amplification plot review or if repeat testing was done. If no repeat testing was done, why are two Ct values reported for Hu18S Ct (b) (4) for sample (b) (6) and (b) (4) for sample (b) (6)?*
- b. *Please clarify if for each donor sample along with Ct value, the amplification curve is analyzed by the analyst irrespective of negative or positive result.*

Oxford Immunotec response to question #9

- a. The sponsor explained that the final result was based on initial testing and amplification plot review. However, the (b) (4) software requires that any sample with a *Babesia microti* Ct value also show a repeat entry to be able to complete the test reporting routine. Even though the curve was not exponential, samples were repeated to satisfy the software requirement so that the negative results could be reported to the blood center. SOP LAB-MOL-BPCR-10, *Babesia microti* Detection

by Nucleic Acid Test for Use on Blood Donor Screening Samples, (Attachment 9.1) has been updated to clarify which samples need repeat testing.

- b.* Oxford Immunotec indicated that analysts are responsible for evaluating each result for interpretation and acceptability. To confirm this has been done, the analyst signs the printed amplification plot which is also reviewed by lab management before results are generated.

The responses are acceptable.

FDA Question #10

In your NAT Amendment response received December 14, 2016 to FDA question 8b (IR dated Nov 11, 2016) for the sample (b) (6), you have stated that “The initial curve was noted as a real, exponential curve. This result could not be repeated, resulting in the specimen report being “Inconclusive”. You stated that the result could not be repeated, but still repeat testing results are reported in the document (MSTDONOR_PROSPECTIVE (Attachment-2.2_MSTDONOR_BCR-NAT-ATT-6)). Please clarify this discrepancy.

Oxford Immunotec response to question #10

The sponsor clarified that when stating that “the result could not be repeated” they should have explained that the positive result could not be repeated since all repeat testing was negative. Per SOP LAB-MOL-BPCR-10, (Attachment 9.1) Section 10.6.1.1.2, if a sample is initially positive but none of the 3 repeats are positive, the specimen is reported as “inconclusive”.

The response is acceptable.

FDA Question #11

In your NAT Amendment response received December 14, 2016 to FDA question #13 (CR dated September 29, 2015) regarding stability studies, you have submitted overview of stability studies. Please clarify the following regarding these studies:

- a.* You have stated that “LAB-MOL-BPCR-7 has been obsoleted and replaced with more robust and complete documents”. Please submit the current SOP that is in compliance with your quality systems (dated, signed, version # etc.).
- b.* Please provide the results obtained for stability analysis after November 2016, if available.
- c.* Please clarify which “finished device lots” were used in these studies.
- d.* Please submit the following documents: DOC-RPT-63; DOC-STB-RPT-25; DOC-STB-25. Please ensure that these documents are signed and dated.

Oxford Immunotec response to question #11

- a. The sponsor indicated that the documents that have replaced the obsolete LAB-MOL-BPCR-7 are summarized in Table 11.1, page 2. See the “Document ID” column for a list of the current SOPs that were created to replace LAB-MOL-BPCR-7 and the “Attachment Number” column for their corresponding attachment numbers.
- b. Tables 11.3, 11.4, 11.5 and 11.6, pages 3-9, show the stability results obtained after November 2016 for the *B. microti* NAT system components. All results are shown as the averages of Ct values.
- c. The Finished Device Lots used are (b) (4).
- d. The information is included in Table 11.1, page 2.

The sponsor provided the information requested by FDA. The responses are acceptable.

FDA Question #12

In your NAT Amendment response received December 14, 2016 to FDA question #18 (CR dated September 29, 2015) on finished device and lot, you have clarified the components of a lot; please provide further clarification regarding which components of the finished device are the critical components.

Oxford Immunotec response to question #12

The sponsor clarified that all components of the NAT Finished Device Lot constitute critical components.

The response is acceptable.

- Chemistry, Manufacturing and Controls

FDA Question #13

In your responses to FDA question #25 on physicochemical acceptance criteria for the purchased oligonucleotides, in Table 25.1 and 25.2 you have stated that the purity requirement for oligo is (b) (4) and for (b) (4) is “pass.” Ideally oligonucleotides used in NAT screening assays are (b) (4) pure. Please clarify the acceptance criteria for the purity of the oligonucleotides. Additionally, the requirement for (b) (4) is a “pass” result from the contract manufacturer (b) (4) of the target calculated (b) (4). In documents submitted in attachment 25.1, the COA doesn’t have (b) (4) results (peak) from the contract manufacturer. Please clarify how the physiochemical characteristics of the primers and probes manufactured by contract manufacturer verified.

Oxford Immunotec response to question #13

The sponsor explained that the purity requirement for oligonucleotides is (b) (4) by (b) (4) according to the manufacturer. If this criterion is not met, the product is not released to customers. Oxford Immunotec verified this information during the vendor audit on January 10th, 2017 and will request a copy of the (b) (4) raw data for each delivered lot of oligonucleotides going forward.

The response is acceptable.

III. Letter Ready Comments

1. In your NAT Amendment response received on December 14, 2016 to FDA question #8 (CR dated September 29, 2015), regarding the analytical specificity/cross reactivity study, you have submitted new data that includes testing of *Plasmodium sp.*, *Trypanosoma cruzi*, and *Borrelia burgdorferi*. The results indicate that none of these pathogens show cross-reactivity in your assay. However, you did not include *Leishmania sp.* infected blood.

Please make all possible efforts to include *Leishmania sp.* DNA samples to evaluate the specificity of the test. If you can't find *Leishmania sp.* infected blood, FDA suggests you use commercially available DNA to spike normal human blood.