



Our Reference: BLA 125685/0/55
CRMTS # 12374

MEETING SUMMARY

April 15, 2020

Enzyvant Therapeutics Inc.
Attention: Dr. Kevin Healy
c/o 324 Blackwell Street, Suite 1220
Durham, NC 27701

Dear Dr. Healy:

Attached is a copy of the memorandum summarizing your March 19, 2020, Type A teleconference with CBER. This memorandum constitutes the official record of the teleconference. If your understanding of the teleconference outcomes differs from those expressed in this summary, it is your responsibility to communicate with CBER as soon as possible.

Please include a reference to BLA 125685/0/55 Meeting ID# 12374 submissions related to the subject product.

If you have any questions, please contact Adriane Fisher at (301) 796-9691.

Sincerely,

Nannette V.
Cagungan -S

Digital signed by Nannette V. Cagungan -S
DN: c=US, o=U.S. Government, ou=HHS, ou=FDA,
ou=People, 0.9.2342.19200300.100.1.1=130018035,
cn=Nannette V. Cagungan -S
Date: 2020.04.15 14:17:05 -0400'

Nannette Cagungan, MS, PD, RAC
Branch Chief
Division of Regulatory Project Management
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Center for Biologics Evaluation and Research

Meeting Summary
(Includes Preliminary Meeting Responses)

Meeting ID #: CRMTS # 12374
Submission type & #: BLA 125865/0/55
Product name: Allogeneic processed thymus tissue-agdc [RETHYMIC]
Proposed indication: For the immune reconstitution of pediatric patients with congenital athymia
Applicant: Enzyvant Therapeutics
Meeting type: Type A
Meeting category: OBLA/CMC
Meeting date & time: March 19, 2020 14:00 -15:00 pm, EST
Meeting format: Teleconference
Meeting Chair: Thomas Finn, PhD
RPM/Meeting Recorder: Adriane Fisher, MPH, MBA
Preliminary Meeting Responses: March 17, 2020

FDA Attendees:

Ekaterina Allen, PhD, OCBQ/DMPQ
Marie Anderson, MS, PhD, OCBQ/DBSQC
Qiao Bobo, PhD, OCBQ/DMPQ
Suzanne M. Carter, OCBQ/DBSQC
Dennis Cato, BS, OCBQ/DIS
Christine Drabick, MS, OCBQ/DIS
Jaikumar Duraiswamy, PhD, OTAT/DCGT
Melanie Eacho, PhD, OTAT/DCGT
John A. Eltermann, Jr., RPh, MS
Thomas Finn, PhD, OTAT/DCGT
Adriane Fisher, MPH, MBA, OTAT/DRPM
Andrea Gray, PhD, OTAT/DCGT
Lily Koo, PhD, OCBQ/DMPQ
Randa Melhem, PhD, OCBQ/DMPQ
Laura Ricles, PhD, OTAT/DCGT
Cong Wang, PhD, OBE/DB/TEB
Zhenzhen Xu, PhD, OBE/DB
Carolyn Yong, PhD, CBER/OTAT
Boguang Zhen, PhD, OBE/DB

Applicant Attendees:

(b) (4), MD, PhD, Professor of Pathology, (b) (4)
Kevin Healy, PhD, Vice President Regulatory Affairs and Quality, Enzyvant Therapeutics, Inc.
Andrea Ashford-Hicks, Senior Vice President, T-Cell Platform Head, Enzyvant Therapeutics, Inc.
Rachelle Jacques, CEO, Enzyvant Therapeutics, Inc.
(b) (4), MD, Director, (b) (4)
(b) (4)
(b) (4), MD, PhD, Professor of Pediatrics and Immunology, (b) (4)

(b) (4), PhD, Vice President, Head of CMC Biologics and Gene Therapy, (b) (4)

(b) (4), PhD, Director of Regulatory Affairs and Quality, (b) (4)
Karin Pihel, PhD, Director, CMC Regulatory Affairs, Enzyvant Therapeutics, Inc.
Timea Pruner, MS, Director Quality, Enzyvant Therapeutics, Inc.

(b) (4), (b) (4) Laboratory Manager, (b) (4)

Background and Objectives:

Applicant submitted a meeting request on February 13, 2020, to discuss with FDA on a clear and expedient path to resolve the deficiencies noted in the complete response letter (CRL), December 4, 2019, and consequently obtain FDA approval of RETHYMIC. Enzyvant intends to address every item listed in the CRL and is fully committed to increasing the rigor of its (b) (4) operations. The pre-meeting materials were submitted on February 13, 2020.

FDA provided its preliminary meeting responses to Enzyvant Therapeutics questions on March 17, 2020. After reviewing the preliminary meeting responses, Enzyvant Therapeutics notified FDA on March 18, 2020, of its decision to limit the meeting to discuss only questions 17,14,3,4,5, and 13.

Preliminary Meeting Responses

We have tried to provide comprehensive responses to all of the questions you posed. We have reviewed the information in your briefing package, previous responses submitted as amendments to the BLA during the original submission review period, multiple versions of referenced standard operating procedures (SOPs) and forms, inspection discussion items, relevant sections of your BLA, and Mid and Late Cycle Meeting correspondence as the basis for our responses. A more comprehensive review will be performed on the information you provide in your BLA resubmission. Therefore, you should consider the Agency's responses as a general perspective and not a definitive assessment of the adequacy of your responses to complete response letter (CRL) items.

Applicant Question 1: Overall, does the Agency agree that the approach submitted on 14 October (Seq 0043) and 31 October (Seq 0050), combined with the information planned for inclusion in the BLA resubmission as described below, will adequately address the Agency concerns and may satisfactorily resolve the outstanding PLI issues?

FDA Preliminary Meeting Response to Applicant Question 1:

We appreciate efforts you have made to respond to specific 483 inspection observations, revisions to your quality system, and updates to your facility design. However, the 483 information provided in eCTD seq 0043 and seq 0050 and your meeting package (seq 0057) are insufficient to completely address this question. Please see our responses to Q.2-12 for specific feedback. Please note that as part of responding to the complete response letter items, it will be important for you to demonstrate that you have an adequate quality system in place and provide confidence in your ability to mitigate risk to appropriately manage manufacturing changes. In your resubmission, please be sure to provide information on your enhanced quality system and list any corrective actions you have taken as a consequence.

Meeting Discussion for Applicant Question 1:

There was no discussion of this question during the meeting.

Applicant Question 2: Does the Agency agree that submission of the SOP updates in the BLA resubmission will address the Agency request to submit documentation demonstrating that PLI Observation 1 has been adequately resolved?

FDA Preliminary Meeting Response to Applicant Question 2:

The proposed changes are acceptable if properly executed and enforced. However, the information you submitted does not include corrective actions taken to address the existing deficiencies, which include: 1) opening deviations for not following existing CAPA SOP to document implemented CAPA and effectiveness evaluation, 2) conducting re-evaluation of all previous deviations and CAPAs to identify and address deficiencies, and 3) opening CAPAs based on the gap analysis results. Further, numerous previous deviation investigation reports did not document whether specific risks were considered, provided sufficient justification for how likely such an event might occur in the future, or fully consider if revised procedures were needed. In addition, with the previous risk matrix scoring system, potential harm to a patient could have been deemed an optional corrective action. For example, according to SOP COMM-QA-077, something that resulted in or could have resulted in transient or persistent medical reaction or injury that is not life threatening but required monitoring and/or intervention to prevent harm, or if the event resulted in or contributed to death or could have resulted in death, but the probability of it happening again was low, then corrective action was recommended, but not required. In your BLA resubmission please clearly indicate how your revised investigation of deviations and requirements for corrective action adequately protect product quality and the patient.

Meeting Discussion for Applicant Question 2:

There was no discussion of this question during the meeting.

Applicant Question 3: Does the Agency agree that the quality system assessments included in PLI Information Amendment 2 on 31 October 2019, along with an update in the BLA resubmission summarizing all completed actions, will address the Agency request to submit documentation demonstrating that PLI Observation 2 has been adequately resolved?

FDA Preliminary Meeting Response to Applicant Question 3:

The changes appear to indicate an improved process; however, whether this addresses all of our concerns will depend on the extent of modifications to your quality system, and when these will be fully implemented. Also, the changes to your quality system are significant, and thus it would be advisable that you review all deviations associated with RVT-802 lots since the process was transferred to the (b) (4) facility in 2016. If you perform a retrospective review, please provide a list of any corrective actions you have taken as a consequence, along with a summary of your risk assessment for each. Further, you have not provided much information in your BLA on the multiproduct nature of your facility and steps taken to minimize impact to the commercial product from other products made in the same facility or using shared resources. Please take this into consideration as you revise your quality system and risk management. As you revise your quality system, we recommend you consider the following:

1. You rely on histology for many aspects of assessing adequate product quality for release. You indicate that you have no scientific or medical justification to set limits. Please note that the purpose of establishing release criteria is to decide whether a product lot is acceptable for patient treatment.
2. In 2016, you switched from using (b) (4) for histology. Each method has its strength and weakness, but one advantage of (b) (4) of the section. However, because of the difference in methodology, you now state that no direct comparison of lots manufactured for your safety and efficacy data sets can be made. This should have been a consideration in switching methods. In your BLA resubmission, please document that the same conclusion can be drawn as to adequate product quality for release using either (b) (4) for histology.
3. There is inconsistency in your appraisal of the value of each tissue slice. For example, we note the following:
 - a. In your package insert you state that as many slices as possible should be transplanted. You also have reservations about sacrificing an additional slice for testing because you consider each slice important. However, regardless of the amount of tissue collected there has been no requirement to target either the average number of slices patients received under IND, or the maximum specified by protocol.
 - b. You do not monitor slice thickness or slice size beyond the required (b) (4) of the filter at Day (b) (4) (which is not verified), nor is there an effort to monitor (b) (4). You have indicated you do not believe these parameters are critical, as there is a lack of correlation between product dose or the number of slices transplanted and clinical outcome. Please note that a lack of a correlation does not necessarily indicate that these parameters are not important.
 - c. You also state that large errors associated with calculating the surface area of small slices are not consequential because the small slices make up a minor part of the total. If small slices represent so little of the total surface area it is not clear why they are included for transplant.

We suggest you make a decision about the relative value of each slice and make any necessary adjustments to manufacturing, testing, and labeling accordingly.

Meeting Discussion for Applicant Question 3:

Enzyvant sought clarification on FDA responses 1 and 3b to Question 3. Regarding justification for setting limits for adequate product quality by histology, Enzyvant indicated they believe their current specifications are justified, as previously documented in Section 3.2.S.4.5. FDA responded that justification for histology acceptance criteria was difficult to determine due to a lack of histology data on clinical lots used to support safety and efficacy. Lot release testing should exist to determine if a product lot is of adequate quality for patient treatment. It would be very helpful to see histology data on clinical lots so that a comparison can be made with the assay validation data.

Regarding the FDA response 3b that large errors associated with calculating the surface area of small slices are not consequential because the small slices make up a minor part of the total, Enzyvant felt the assay was validated to show acceptable accuracy of the surface area measurement and the validation was provided in the BLA. FDA response that for the accuracy of the surface area calculation for product dose, Enzyvant had established an allowable variance of (b) (4) for the assay. The data provided on the (b) (4) measurements showed that errors associated with (b) (4) thresholds according to SOP versus thresholds on individual slices could be as large as (b) (4) for some slices. Though variability across all slices for a product lot fell within (b) (4), this was just for one variable. Assay validation normally takes into consideration different analysts, repeat testing by the same analyst, different lots, etc. All product assays have some level of assay variability, but typically known sources of variability that can be controlled are usually optimized to reduce variability to the extent feasible. FDA recommend that Enzyvant also consider the impact of (b) (4) variability. For example, Enzyvant should consider situations where a patient had already been treated with RATGAM, but the product surface area was close to the minimum. Enzyvant indicated they would perform a reassessment of the assay but are not planning on revalidating the assay.

Applicant Question 4: Does the Agency agree that the approach described in the (b) (4) PLI Information Amendment, and an update summarizing the completed actions in the BLA resubmission including the EMPQ report, modifications to the EM program, and a summary of the updated routine EM program, will adequately address the Agency request to submit documentation demonstrating that PLI Observation 3 has been adequately resolved?

FDA Preliminary Meeting Response to Applicant Question 4:

The reports you provided do not address all the deficiencies identified during the inspection regarding EMPQ, routine monitoring under (b) (4) conditions, differential pressure/temperature/humidity monitoring, and associated acceptance criteria. Also, the risk assessment used for preparing the EMPQ protocol includes several inconsistencies that require explanation and justifications.

Furthermore, you intend to implement facility modifications, procedural modifications, and modified flows. Please note that the submitted risk assessment, the worst-case EM locations selected for the pre-change EMPQ, and the associated EMPQ results are not expected to be representative of the post-change facility. Therefore, once the changes have been implemented, another risk assessment should be performed to determine the worst-case EM locations, and a new EMPQ should be completed to qualify the post-change facility. As such, in your response to the CRL, please include a description of the implemented changes, a new risk assessment for EM monitoring, a new EMPQ (b) (4) conditions), and routine monitoring (b) (4) conditions) and trending report for the classified areas to ensure they meet the acceptance criteria.

Meeting Discussion for Applicant Question 4:

(b) (4)/Enzyvant thanked the FDA for the comments and explained that a new risk assessment and EMPQ will be performed after the implementation of the facility modifications.

They also requested clarifications about the inconsistencies that were noted in the risk assessment.

FDA mentioned that clarification is needed regarding the following items noted during review of the October 2019 EMPQ response (Table 9) and the risk assessment Appendix 1 (b) (4)-2019-045-p):

Items that need clarification in Table 9:

- You stated the pre-EMPQ is performed under (b) (4) conditions and EMPQ under (b) (4) conditions, but you did not specify the status of Post-EMPQ (p.16). Please note FDA's expectation of environmental monitoring to be performed under (b) (4) conditions as they are representative of the manufacturing operations.
- You reported that personnel monitoring is performed in all rooms (p.17). Clarify how and where.
- You stated that (b) (4) are monitored for the incubators are performed (b) (4) (p.17). However, in the risk assessment (b) (4)-2019-045-p) it is stated that only (b) (4) monitoring of incubators is performed (p.14).
- Pressure differential: The frequency of monitoring is not clear. The EMPQ report narrative states that it is recorded (b) (4) between the ISO (b) (4) and ISO (b) (4) and between the ISO (b) (4) and CNC. However, in Table 9 (p.17) it is stated that the pressure is (b) (4) monitored in all rooms.

Items that need clarification in Appendix 1 (b) (4)-2019-045-p):

- The (b) (4) is sampled for (b) (4) at the (b) (4) of the (b) (4) (p.11); yet that would not be possible during manufacturing, and is not what we observed on inspection where the (b) (4) monitor was placed to the (b) (4) of the (b) (4), and the (b) (4)
- Several entries in Appendix 1 regarding the cleaning activities:
 - Yes (not specifically designated per SOP)
 - Yes (not performed)
 - The frequency of cleaning: (b) (4). Many areas are not assigned the (b) (4) cleaning, even though those areas are supposed to be cleaned (b) (4) as stated in (b) (4)-SOP-006.

(b) (4)/Enzyvant stated that they identified some of the inconsistencies after the submission of the documents and would work on reviewing and revising the documents accordingly. Enzyvant requested that FDA include the mentioned examples in the meeting minutes.

Applicant Question 5: Does the Agency agree that the approach described above is reasonable to address Observation 4? Does the Agency agree that an update in the BLA resubmission summarizing the completed actions, including the DE study report and documentation of the revised cleaning program, will address the Agency request to submit documentation demonstrating that PLI Observation 4 has been adequately resolved?

FDA Preliminary Meeting Response to Applicant Question 5:

The information provided in the meeting package, including the SOPs, does not contain sufficient details to ensure proper disinfectant validation and appropriate routine cleaning. Furthermore, you stated that the cleaning procedures for the facility will be revised based on EMPQ results and revised again after the planned facility modifications. As such, we cannot comment on the sufficiency of the information you have provided thus far in addressing Observations 4. However, we have the following comments for your consideration:

1. Please provide the cleaning program of the facility after the proposed facility/procedural changes have been implemented.
2. You reported that the DE study protocol was modified to accommodate changes made to the coupon surfaces. However, insufficient details were provided to support your evaluation and justify all the changes made to the surface coupons. In the DE study protocol, please list all the facility surfaces. If you intend to group surfaces with similar characteristics, please identify the worst-case representative for each group based on your risk assessment (e.g., characteristics, usage in facility, etc.).
3. It appears that you plan to change one or more cleaning agents. Please ensure the new cleaning agent(s) is/are validated and implemented for routine use prior to performing the post-change EMPQ. Alternatively, present your risk assessment and supporting data to demonstrate that the new cleaning agent(s) is/are as effective on the facility flora and therefore has/have no adverse impact on the existing EM program.
4. The DE studies should be performed using representative/worst-case soils, and should cover all facility surfaces (i.e., not the most probable surfaces that would be soiled).
5. Additional information and justification are needed to support the cleaning frequency, the disinfectants used, and demonstration of their effectiveness on
6. Please provide information to support the following concerns in your response to the CRL:
 - a. Rationale for why some surfaces are being exposed to soil and microorganisms, while others are being exposed to microorganisms only
 - b. Rationale for (b) (4) rotation of disinfectants (b) (4) for (b) (4) facility cleaning/disinfection. These (b) (4) disinfectants are not active on the same microorganisms based on the information you have provided.

- c. Rationale for not including (b) (4) in the DE studies, considering it is widely used in the facility.
- d. Rationale for selecting a representative surface for each category of surface materials.
- e. Rationale for listing (b) (4) specific facility isolates, and then stating that (b) (4) currently has no facility isolates.
- f. Rationale for not using all recommended compendial aerobic isolates.

Meeting Discussion for Applicant Question 5:

(b) (4)/Enzyvant stated that they are considering several protocol modifications to address the FDA comments including adding (b) (4) to the DE study for the bacterial and fungal challenge. They explained that the DE study was designed to evaluate disinfectant efficacy on all facility surfaces, with the soiled arm of the study focused on facility surfaces that are in closest proximity to open product. The product is only open in the (b) (4) and therefore (b) (4) surfaces are appropriate for the soiled arm. Spills have never been observed in the incubator. To address the Agency comment #4, the protocol can be modified to expand the soiled arm to include (b) (4), which are materials present in the (b) (4). The remaining facility surfaces outside of the (b) (4) and not in close proximity to open product (b) (4) are not planned for inclusion in the soiled arm but are included in environmental monitoring.

FDA responded that the DE studies should be performed using representative/ worst-case soils, and should cover all facility surfaces (i.e., not only the most probable surfaces that would be soiled). FDA added that while the proposed study does not include all the facility surfaces, the response meets the immediate concerns. FDA requested that Enzyvant provides, in the BLA resubmission, a list of all facility surfaces (and their locations), and what surfaces (coupons) were subjected to disinfectant effectiveness as worst-case surface representatives. FDA also requested that the applicant should provide justifications for not including the powder coated steel and epoxy surfaces. FDA stated that the information will be evaluated, and additional feedback will be provided during the BLA resubmission review.

(b) (4)/Enzyvant stated that (b) (4) additional organisms will be included in the testing, such that the standard set of compendial organisms, along with (b) (4) facility isolates, will be tested. FDA agreed with their proposal. FDA requested clarifications about the basis for selecting the facility isolates, even though they do not have a facility isolates program. (b) (4)/Enzyvant clarified that the (b) (4) selected isolates ((b) (4)) were identified during environmental monitoring. FDA recommended that (b) (4)/Enzyvant should have a program to evaluate the facility isolates as the flora changes with seasons and activities.

Applicant Question 6: Does the Agency agree that an update in the BLA resubmission summarizing the completed actions, including the information described above, will address the Agency request to submit documentation demonstrating that PLI Observation 5 has been adequately resolved?

FDA Preliminary Meeting Response to Applicant Question 6:

The 483 Observation 5 response and the meeting package are not detailed enough to determine their adequacy. The following should be provided for our review:

- Completed assessment of the (b) (4) alarm systems, with the outcomes
- IQ/OQ of the non-qualified (b) (4) alarms
- Red-lined versions of updated SOPs CT2-SOP-094, CCBB-LAB-010, and any other SOPs detailing (b) (4) installation/replacement (including selecting location for sensor placement), qualification, and preventative maintenance, alarm setup, periodic data review [scope, frequency, and responsibility, excursion handling (including product impact assessment)].

Additionally, please address the following issues:

1. No action was specified regarding existing sensors, where equipment mapping data is already available. No commitment was made to move existing probes to the worst-case locations identified during the mapping studies.
2. Both existing and new (b) (4) alarms should be included in your preventive maintenance program; required preventive maintenance should be specified.
3. (b) (4) data should be reviewed periodically, in addition to just alarms, to ensure continuous system functionality. The data review frequency should be based on criticality of the equipment.
4. Your 483 response stated that (b) (4) Quality review of the (b) (4) data is performed (b) (4) per CCBB-LAB-010. During the inspection, it was discovered that CCBB-LAB-010 does not require (b) (4) data review. (b) (4) staff monitors (b) (4) data and responds to the alarms. Relaying this information to (b) (4) quality is not proceduralized.
5. Please explain what would be considered adequate alarm notification and response for various types of equipment used. Please clarify what studies, if any, will be performed and what existing data would be used to support setup of the alarms.

Please specify whether the additional differential pressure sensors you plan to install will be continuously recording and connected to an alarm.

Meeting Discussion for Applicant Question 6:

There was no discussion of this question during the meeting.

Applicant Question 7: *Does the Agency agree that the procedures and assessments submitted on (b) (4) and an update summarizing the completed actions in the BLA resubmission, including the information described above, will address the Agency request to submit documentation demonstrating that PLI Observation 6 has been adequately resolved?*

FDA Preliminary Meeting Response to Applicant Question 7:

Regarding Observation 6a: The proposed changes are acceptable if properly executed and enforced.

Regarding Observation 6b: The revised procedures represent significant improvements, but we recommend a careful review of how process step times and hold times are defined and justified. For example, step (b) (4) on form (b) (4)-SOP-029-FRM1 (both original and revised) states that the tissue must be processed within (b) (4) of notification, implying that all slicing of the tissue must be complete within (b) (4). The revised (b) (4)-SOP-029-FRM2 now includes a place to calculate and document the elapsed hold time, but the (b) (4) is based on the *start of thymus processing*, and thus the two forms are inconsistent. Steps (b) (4)-SOP-029-FRM2 involve preparing the (b) (4), instrument, dishes, labels, and slicer for use. Actual tissue processing does not begin until step (b) (4) and the time is not recorded for step (b) (4). Since the time for Step (b) (4) was not documented it is unclear for lot (b) (4) whether the tissue had expired prior to the initiating of processing. If processing was supposed to be complete, it appears the source material had expired before it was finished. Further, it is unclear for a lot that was used to treat a patient why (b) (4) storage was necessary given the time of notification was (b) (4). Prior to conducting process validation lots please be sure to update documents appropriately and train individuals accordingly. You may wish to consider adding definitions of specific terms to your SOPs and processing forms to avoid any confusion.

It is also unclear why processing tissues was initiated near the (b) (4) expiry the next day and was not expedited. According to Table 6 of your briefing document, notification for lot # (b) (4) was at (b) (4), yet (b) (4) storage was used, and the processing began nearly (b) (4) later. On inspection it was indicated that (b) (4) storage is used in cases where arrival of the tissue into the facility is due to delay by the OR and the tissue is not available until late in the afternoon. One of the justifications for the original (b) (4) proposed total tissue hold time at room temperature was only a single manufacturing suite (room (b) (4) is available for all RVT-802 activities, including media preparation. If the room is busy the tissue may have to sit at room temperature. It is not clear how manufacturing suite time is prioritized and whether adequate resources are available to handle the full proposed manufacturing scale of (b) (4) lots being produced within the same room over the course of up to 21 days with (b) (4) media changes. In your BLA resubmission please include information in Section 3.2.R on manufacturing capacity, taking into consideration the logistics of manufacturing steps and hold times.

It is also unclear why processing tissues was initiated near the (b) (4) expiry the next day and was not expedited. According to Table 6 of your briefing document, notification for lot # (b) (4) was at (b) (4), yet

(b) (4) storage was used, and the processing began nearly (b) (4) later. On inspection it was indicated that (b) (4) storage is used in cases where arrival of the tissue into the facility is due to delay by the OR and the tissue is not available until late in the afternoon. One of the justifications for the original (b) (4) proposed total tissue hold time at room temperature was only a single manufacturing suite (room (b) (4) is available for all RVT-802 activities, including media preparation. If the room is busy the tissue may have to sit at room temperature. It is not clear how manufacturing suite time is prioritized and whether adequate resources are available to handle the full proposed manufacturing scale of (b) (4) lots being produced within the same room over the course of up to 21 days with (b) (4) media changes. In your BLA resubmission please include information in Section 3.2.R on manufacturing capacity, taking into consideration the logistics of manufacturing steps and hold times.

Meeting Discussion for Applicant Question 7:

There was no discussion of this question during the meeting.

Applicant Question 8: *Does the Agency agree that the submission of the PLI Information Amendment on (b) (4), and an update summarizing the completed actions, as described above, in the BLA resubmission will address the Agency request to submit documentation demonstrating that PLI Observation 7 has been adequately resolved?*

FDA Preliminary Meeting Response to Applicant Question 8:

The updated (b) (4)-SOP-060 states that a certification is performed every (b) (4) by an approved vendor (b) (4) using standards traceable to (b) (4). Section states “Any leaks found must be repaired before the (b) (4) is returned to use. Leak will be repaired with (b) (4). Repair of HEPA filters should be restricted to limited area before complete filter replacement is required and such limit should be established in the SOP. In addition, the appropriateness of the acceptance criteria for (b) (4) should be verified through aseptic process simulation, (b) (4) studies related to the thymus tissue processing, operator qualification, and other process validation activities.

Meeting Discussion for Applicant Question 8:

There was no discussion of this question during the meeting.

Applicant Question 9: *Enzyvant and (b) (4) are executing these studies; summaries and data will be included in the BLA resubmission. Does the Agency agree that summarizing the completed actions in the BLA resubmission, including the information described above (but exclusive of the realtime aging study if it is not completed), will address the Agency request to submit documentation demonstrating that PLI Observation 8 has been adequately resolved?*

FDA Preliminary Meeting Response to Applicant Question 9:

Regarding Item 8a: It is unclear if the revisions to SOP COMM-QA-019 Change Control and your gap analysis would prevent future occurrences of changes being implemented before appropriate oversight had been completed.

Regarding Item 8b: We cannot comment on the adequacy of your response until the

following issues are addressed:

1. The endotoxin acceptance criterion for your ancillary materials of (b) (4) is not appropriate because the limit is intended for a single implanted device and does not take into account the cumulative endotoxin contribution from each of the ancillary material to the final product.
2. Given that the test materials are not liquid and different buffer volumes might be required for endotoxin recovery from the articles, a per item specification should be included for all materials.
3. Your corrective action does not include updating sterile and endotoxin-free supply vendor qualification.
4. You proposed to reduce supply testing frequency to (b) (4) upon acceptable testing of (b) (4) lots. Please clarify the course of action if one or more of (b) (4) tested lots fail to meet the acceptance criteria (e.g., retesting of failed lots and/or impact on vendor qualification and testing reduction for future lots).
5. You proposed to release (b) (4) based on sterility results for (b) (4) as all of these materials are sterilized using (b) (4). Please note that such strategy requires the items to be (b) (4) in the same load.
6. Regarding SOP (b) (4)-GEN-009, Assigning Lot Numbers and Expiration Dates to Reagents and Materials, please ensure it covers assigning expiry dates to sterile materials that are not covered by (b) (4) testing and have no expiry date provided by the manufacturer.
7. Please see additional comments regarding (b) (4) container below (response to Q. 19).

Regarding Item 8c: The proposed identity tests are acceptable. In your BLA submission please provide copies of identity results for each ancillary material and assay validation reports.

Regarding Item 8e: Given the reliance on the vendor shelf life studies, you should include review of the study reports in your vendor qualification procedures.

Meeting Discussion for Applicant Question 9:

There was no discussion of this question during the meeting.

Applicant Question 10: Does the Agency agree that an update in the BLA resubmission summarizing all of the completed actions, including the information described above, will address the Agency request to submit documentation demonstrating that PLI Observation 9 has been adequately resolved?

FDA Preliminary Meeting Response to Applicant Question 10:

Regarding Item 9a-c: Your responses are acceptable. Please ensure you submit redlined versions of SOPs and/or previously submitted documents. Additionally, in your response to CRL, please include the following information:

- Describe the use of (b) (4) Storage Current Inventory” form.
- Clarify discrepancies between room temperature and (b) (4) storage logs; it
- Appears that the same log is used for adding and removing cold storage supplies whereas ambient storage supplies are added and removed on two separate logs.

Regarding Item 9d: Information provided in your (b) (4) , additional responses to 483 observations (serial #50) are acceptable. We cannot comment on planned changes listed in your briefing document because insufficient detail was provided. In your BLA resubmission please also include information on who will have access to (b) (4) sample storage areas and (b) (4) , especially considering research samples are stored in the same (b) (4).

Meeting Discussion for Applicant Question 10:

There was no discussion of this question during the meeting.

Applicant Question 11: Does the Agency agree that an update in the BLA resubmission summarizing all of the completed actions, including the information described above, will address the Agency request to submit documentation demonstrating that PLI Observation 10 has been adequately resolved?

FDA Preliminary Meeting Response to Applicant Question 11:

The information provided is considered deficient for the following reasons: 1) The (b) (4) schedule for data backup appears insufficient to protect against data loss during the (b) (4) period; 2) It is not clear what data protection measures, including audit trail requirements, are in place for the (b) (4) computer; 3) It is not clear what data protection measures, including audit trail, are in place for the removable (b) (4) ; and 4) it is not clear if the PDF report of the sterility data represents a true copy of the original data, including any associated metadata, which should be compatible with the original format to allow data recovery. Please address the concerns or implement a new data backup procedure supported by a new validation study.

In your BLA resubmission please provide a table of all software used in manufacturing and testing, and a list of the types of digital files saved for each lot. Please include a summary of who has access to the files and how the files are backed up.

Meeting Discussion for Applicant Question 11:

There was no discussion of this question during the meeting.

Applicant Question 12: Does the Agency agree with this planned approach to address Observation 11? Does the Agency agree that an update in the BLA resubmission summarizing the completed actions, including the information described above, will address the Agency request to submit documentation demonstrating that PLI Observation 11 has been adequately resolved?

FDA Preliminary Meeting Response to Applicant Question 12:

In your proposed risk assessment to identify any gaps in the current equipment performance qualification (PQ) and preventative maintenance (PM) programs, please consider the Agency’s comments and concerns communicated through inspection

discussions and 483 Observation citations (e.g., pre-specified PQ/PM acceptance criteria that are relevant to product quality, validated parameters being applied to the routine manufacturing process and incorporated in relevant SOPs, identified critical process parameters being monitored/trended and documented in batch records, etc.). Additional PQ activities may be required based on your risk analysis outcomes and should be completed prior to new PPQ protocol development and execution.

Meeting Discussion for Applicant Question 12:

There was no discussion of this question during the meeting.

Applicant Question 13: *Does the Agency agree that implementation of the modifications proposed above will resolve this deficiency? Specifically, does the Agency agree with Enzyvant's plans to conduct in-process (b) (4) testing on days (b) (4) of culture, respectively, and histology testing on another slice taken from the (b) (4) days before the implant surgery?*

FDA Preliminary Meeting Response to Applicant Question 13:

The approach is generally acceptable but would benefit from further revisions. We have the following recommendations:

Histology testing for release on the (b) (4) safety and efficacy lots reported in Module 3 was conducted on product samples collected (b) (4) days prior to release, with the average being (b) (4) days. At the Late Cycle Meeting you indicated that you typically allow (b) (4) days to obtain histology results, and that depending on the day the samples are collected in the work week, it may take longer. Your proposal to retest beyond (b) (4) days is therefore reasonably consistent. However, you have also indicated that all histology samples for this product are rushed to obtain results as fast as possible. For lots (b) (4) results were obtained the (b) (4) day. In the interest of evaluating samples that most closely resemble what will actually be transplanted into the patient, we recommend that you set a target for sample collection at 2 days prior to release, with an allowance of up to 5 days according to your criteria.

While we agree that Day (b) (4) is a reasonable time period to evaluate the necessary reduction in donor thymocytes in the tissue slices because the largest reduction occurs within the first (b) (4) days, it is not clear that testing at Day (b) (4) is the best approach.

Initiation of RATGAM treatment is (b) (4) days prior to transplant and the earliest a product lot can be released is Day 12. This would mean any time Day (b) (4) samples are collected, repeat (b) (4) testing will be necessary, and could occur as early as Day (b) (4), or as late as Day (b) (4). It is not clear there would be much to gain by retesting on Day (b) (4) whereas testing closer to the day of release (i.e., Day (b) (4)) would be more meaningful. Further, testing at Day (b) (4) for Day 12 release would mean testing for final product release only (b) (4) of the way through the manufacturing process, and some lots undergo significant phenotypic changes between Days (b) (4) 12. We recommend that you establish a retesting table based on the day of intended RATGAM treatment and scheduled transplantation, and choose appropriate target dates that will provide the most meaningful evaluation of final product quality, while still fitting within the (b) (4) day before the implant surgery criteria and the time needed to obtain histology results.

As requested, you will implement upstream testing for product dose to be reasonably certain that prior to patient conditioning with RATGAM that the minimum final product dose can be achieved. You acknowledge that tissue (b) (4) may occur, thus affecting the dose that can be achieved. Data provided in your submission indicates that some cultured slices do (b) (4) during culture, with some (b) (4) substantially. You indicate predicting the level of (b) (4) may be difficult. The specification for (b) (4) dose testing is the same as for final product release. In some cases, the number and size of slices established on Day (b) (4) would likely achieve the minimum final product dose regardless of (b) (4), even for patients of greater body surface area. However, in cases where the number of slices prepared is small, the slice sizes are smaller, or the target patient body surface area is greater (two recent patients were around 2 years of age), the minimum dose may not always be achieved. Given that (b) (4) can occur, and you do not know the exact degree to which the dose could be reduced by further culturing, we would advise including an added margin for interim dose beyond that specified for the final product.

You point out there have been no lots that have failed the endotoxin acceptance criteria. Please note these results were obtained with (b) (4) samples across all dishes that may have (b) (4) endotoxin levels in the test samples below the validated sensitivity level of the assay. While your previous endotoxin results are useful, you should be aware that it is possible you could have endotoxin failures in the future with your revised procedure to (b) (4) medium from no more than (b) (4) dishes.

It is not clear if you plan to retest in the event of a serious product deviation, even if the test window is (b) (4) days.

Meeting Discussion for Applicant Question 13:

Enzyvant sought clarification on FDA's comment about whether a plan was in place to retest the product by histology for release in the event of a serious product deviation, even if the original testing falls within the proposed test window ((b) (4) days). FDA expressed concern that if a serious product deviation occurred after testing was completed within the original (b) (4) day test window, this may be difficult to assess whether adequate product quality remained. An example was provided for a product intended for final harvest and administration on Day 20 that was tested on Day (b) (4) for release, but an incubator failure occurred on Day (b) (4). In such a situation, it is not clear how Enzyvant would judge the impact of the failure on the product, if all histology testing had already been performed.

Applicant Question 14: Does the Agency agree that implementation of the modifications proposed above and provision of the requested data will resolve this deficiency? Specifically:

- *For 3b, does the Agency agree with the proposed approach to implementation of the (b) (4) histology method and acceptance criteria, including use of the qualitative global overall histology assessment?*
- *As noted in 3c, (b) (4) histology samples, which are required for the (b) (4) assay, are not available for most of the subjects with lower naïve T cell counts through Year 1. Does the Agency agree that given the supporting information and Enzyvant's plan to use the (b) (4) assay on 11 subjects, data developed using the Company's*

proposed approach can satisfactorily address this stated deficiency?

FDA Preliminary Meeting Response to Applicant Question 14:

With regards to Item 3a and 3b: The proposed strategy partially addresses our concerns, but is not yet satisfactory as described, specifically:

1. You propose to create an SOP for the histological evaluation of RETHYMIC. Given that the criteria are revised and are (b) (4), please provide reference images for each criterion. Please include the reference images, in addition to clear descriptions why the reference image is assigned a specific (b) (4) criterion. The description should also include a rationale for why the image is being assigned the specific criterion and how it can be discriminated from other images assigned a different criterion within the same parameter. We recommend the use of arrows, boxes, and any other applicable methods of illustration to clearly and definitively specify the tissue hallmarks that enable the pathologist to assign the specific (b) (4) score. The reference images should be provided both for the Agency's review and integrated into the Histology Training Guide, as well as the SOP. To support the use of these reference images, please provide 10 fields of view for the tissue section from which the reference image was obtained. Please note that it is important not only for the pathologist performing the assay to have clear procedures for how the assay is to be conducted and the results generated, but representatives from Quality Control and Quality Assurance who will ultimately have to sign off on those results when making a determination to release the product need to be able to interpret those results. Having clear definitions about what each score means can aid in that process. It is also important for risk management and change control.
2. For each parameter evaluated by the histology assay, please identify the purpose of each parameter and justify how the approach is suitable for the intended purpose.
3. The use of several acceptance criteria does not appear to be in line with the Histology Validation Report provided in the original BLA submission. Specifically, please address the following discrepancies:
 - a. You propose an acceptance criterion of (b) (4) for Day (b) (4) and Day (b) (4) tissue. In the Histology Validation Report, you state, "All thymus tissue samples (b) (4) on (b) (4) were rated as (b) (4)." It is not clear why a Day (b) (4) tissue would have (b) (4) and why this incoming tissue would then be acceptable, as suggested by your acceptance criteria. Please revise the acceptance criteria for (b) (4) to be in line with the validation data or provide a justification for the use of rating of (b) (4) and provide representative examples of each.
 - b. You propose an acceptance criterion of (b) (4) for Day (b) (4) and Day (b) (4) tissue. However, in the Histology Validation Report, all thymus tissues (including Day (b) (4) Day (b) (4), and (b) (4) samples) received a rating of (b) (4). Therefore, it is not clear whether an acceptance criterion of (b) (4) is supported. Please revise the

acceptance criteria for (b) (4) to be in line with the validation data or justify why rating of (b) (4) is supported and provide representative examples of each.

- c. Based on the Histology Validation Report data, all Day (b) (4) tissue received a rating of (b) (4) for the Viability criterion. It is expected that a healthy donor tissue that is properly transported to the manufacturing facility would display (b) (4)

(b) (4) Therefore, please revise the Viability acceptance criterion for Day (b) (4) tissue or provide a justification for allowing incoming tissue that is rated as (b) (4) and provide representative examples of each.

4. For the global histology assessment of Day (b) (4) tissue, you state that the overall tissue morphology is “diagnostic of normal thymus.” Please refine this acceptance criterion to include normal thymus tissue hallmarks that differentiate between normal thymus tissue and abnormal thymus tissue.
5. During the histology method validation, the (b) (4) acceptance criteria were not met in (b) (4) analyses, but you concluded that “the data support using this assay in a (b) (4) fashion.” While we tentatively agree that a method validation will not need to be repeated, we request a justification for the use of (b) (4) acceptance criteria despite the two protocol deviations. Furthermore, in the Histology Method Validation Report for viability, you state, “in cases where the sample is borderline between one rating and another, it is difficult to consistently rate the sample with the same (b) (4) assessment”. The difficulties associated with consistent ratings appear to be, in part, due to “heterogeneity” of (b) (4) samples, which had a lower consistency than the ratings for (b) (4)-stained samples. You state that (b) (4) viability threshold of (b) (4) was “borderline” for the samples that failed acceptance criteria and you have chosen to remove references to percentages for the viability criterion (strikethrough font in Table 4). Please provide a justification for the removal of references to the percentages and refine criteria (b) (4) to further distinguish (b) (4)
- (b) (4) . We recommend defining (b) (4) distributions, and provide reference images for each to ensure that, from a quality control perspective, each parameter is clear.
6. You indicate that by Day (b) (4) there is always a large reduction in thymocytes, though significant levels of (b) (4) cells can persist even out to Day 21. Since it is expected that there will always be a large reduction in thymocytes, we believe a score of (b) (4) for (b) (4) should also reflect a marked T-cell depletion, in addition to the criteria you have established for a score of (b) (4). Strictly interpreted, a score of (b) (4) could be assigned for a tissue section even if no large reduction was seen.
7. With regard to Item 3c: A major challenge in evaluating acceptance criteria for histological analysis for product release in your original submission was the lack of representative images of histology results for the safety and efficacy

data set, which encompasses product lots manufactured as far back as 1993, and histology being used for lot release dating back to 2002. It was difficult to assess the representative images from product development, assay validation, process validation, and stability without knowing the range in assay results from the actual clinical samples. While we recognize that your assay method has changed from (b) (4), the purpose of the assays and the histological features you are looking for to decide on product release are similar. We strongly recommend that you provide examples of histology results from clinical lots used to support safety and efficacy, including product lots where test samples were taken close to the time of culture harvest.

8. According to (b) (4)-SOP-032, (b) (4) samples are (b) (4). However, you state that the (b) (4) histology assay could not be accurately applied to (b) (4) sections due to physical changes induced in tissue components during (b) (4). Please provide a rationale for the currently proposed method for (b) (4) tissue sections for (b) (4). If (b) (4) of sections induces physical changes in tissue components, it is not clear how a retrospective analysis can be performed on future samples.
9. You state that (b) (4) of sections induces changes in tissue components and (b) (4) histology assays cannot be accurately applied to (b) (4) sections. Please provide data from side-by-side comparison from the same tissue processed using the (b) (4) methods (b) (4). Please submit representative fields of view for (b) (4) staining for tissue processed using the (b) (4) methods and a discussion of changes that are induced by (b) (4). Please justify how the change in the methodology will not lead to a different determination of acceptability for release than what was used clinically.
10. You changed the tissue preservation method for (b) (4) samples. Please include a risk assessment for the change in tissue (b) (4) methodology.

Meeting Discussion for Applicant Question 14:

Enzyvant stated they intend to make the adjustments to histology testing suggested by FDA. Enzyvant sought FDA's feedback on their proposal to create a Standard Operating Procedure (SOP) for the histological evaluation. Enzyvant will create an SOP for the histology assays and replace the histology training guide. FDA clarified it was not necessary to discontinue the training guide, especially if the training guide contained information not present in the SOP. Enzyvant explained that the SOP will contain all relevant information from the training guide plus enhanced information on the procedure, and thus is redundant.

Enzyvant removed references to percentages in the batch record collected from histology testing, as was suggested by FDA. Enzyvant explained that the percentages included were not meant to imply a quantitative assessment by histology. To be consistent with the qualitative nature of the method, such percentages will not be estimated in the future. This justification will be provided in the BLA resubmission. FDA understood that these percentages were estimates

and not based on specific numerical counts, but still found the estimated percentages useful, and did not imply that such information needed to be removed. The concern raised during review of the BLA original submission was that the batch record and the histology assessment were capturing information not reflected in the product specifications. Therefore, it is acceptable to include percentage estimates in the records, if this information is useful, and as long as it is consistent with Enzyvant's SOP and specifications. If the proposed 3-point scale adequately captures the same information, that would also be acceptable.

The current SOP is based on over (b) (4) images of tissue at multiple time points from Days (b) (4) to 21, from product lots including (b) (4) clinical lots, (b) (4) previous PV lots and (b) (4) research lots (including (b) (4) lots of (b) (4) samples). These slides were stained with (b) (4) ratings for these images were determined. Enzyvant asked if the number of lots and images is acceptable. FDA indicated that there is no pre-established regulatory expectation for the number of lots or images that would be needed in this case. Enzyvant should justify in their BLA resubmission why they believe the number and types of lots and number of images is sufficient for the intended purpose. Of concern is the number of clinical lots used for analysis and how representative these are of the clinical lots used to support safety and efficacy.

(b) (4) samples of (b) (4) samples are maintained in the Pathology Department. Enzyvant clarified that the samples are (b) (4) instead of (b) (4) to enable more assays to be conducted on (b) (4) tissues. FDA asked which additional tests could be conducted. Enzyvant elaborated the (b) (4) samples could possibly be used for future assay development. Therefore, Enzyvant felt it could be very valuable to have samples for evaluation that were obtained from clinical lots. The Agency agreed (b) (4) samples can be quite useful for assay development, but also emphasized the importance of being able to use such samples for investigation of product deviations, customer complaints, or adverse events. Enzyvant further explained that the tissue preservation method used by (b) (4) since 2001 is similar to that currently used by the (b) (4) facility, and thus no risk assessment is required. FDA indicated a risk assessment should be performed to evaluate any difference in methodology or proposed use. Whether any corrective action or change control is needed would depend on the findings of the assessment, which is typical of risk management. FDA asked if Enzyvant could use the (b) (4) tissue from the clinical lots to provide a more direct comparison of historical lots and recent lots. Enzyvant indicated they could.

As requested, Enzyvant intends to provide a side-by-side comparison of (b) (4) tissues. Enzyvant explained images from (b) (4) slides provide higher quality and would be more likely to lead to rejection of a lot, compared to analysis of (b) (4) tissue. The impact on determination for acceptability for release will be included in the BLA resubmission. FDA acknowledged that (b) (4) generally have superior resolution and that this change was intended to provide an improvement to the histology assay. However, a change in the assay method makes it difficult to make comparisons with product lots intended to support safety and efficacy. The assay method change should have been part of a risk management strategy and discussed in the BLA original submission. FDA stated that one of the most challenging aspects of reviewing the adequacy of product quality in the original

BLA was a lack of histology data on product lots patients received. Without those images to use as a reference, it was very difficult to assess assay validation, PV, and product specification data in order to conclude manufacturing was in an adequate state of control. FDA understands that it may be difficult to apply the exact same histology scale used for (b) (4) with historical (b) (4) tissue section data, but the goal of the assays is the same. It should be possible to make a general comparison of the (b) (4) methods to provide confidence that the new method would lead to the same determination of product quality as the previous method.

FDA also pointed out that since histology assessment is critical to evaluation of multiple critical quality attributes for release, such a change would normally be considered a major manufacturing change. For example, for a commercial product such a change would require a BLA supplement, including a risk assessment and method comparability data. For a quality system change, change management should be implemented.

Applicant Question 15: *Given the information presented above, does the Agency agree that reducing the thymus source material hold time to (b) (4) from the time of notification of tissue availability to start of processing, is appropriate and adequately addresses this deficiency?*

FDA Preliminary Meeting Response to Applicant Question 15:

Yes, we agree the (b) (4) revised hold time proposal is more appropriate given the historical experience. (b) (4) of the (b) (4) lots held at room temperature (with survival 1-year post-treatment) experienced hold times at room temperature for (b) (4) or less. Basing hold times on conditions used for manufacturing clinical lots that have shown positive clinical outcome reduces the reliance on analytical stability data and helps justify the specific values. The proposed tissue hold time should be evaluated in the new PV study to provide additional supportive data. Please note that it is questionable whether tissue held for up to (b) (4) at room temperature prior to processing can be considered to be “processed immediately,” as described under IND 9836 protocols, and underscores the importance of having clear definitions.

Meeting Discussion for Applicant Question 15:

There was no discussion of this question during the meeting.

Applicant Question 16: *In light of the information presented, does the Agency agree that reducing the drug product expiry from (b) (4) resolves this deficiency?*

FDA Preliminary Meeting Response to Applicant Question 16:

The proposed DP expiry appears to be adequate. The proposed DP expiry of (b) (4) aligns with the time out of incubator experienced by DP lots used to examine the tissue hold times. Out of the (b) (4) lots evaluated for the tissue hold times in CR#4, (b) (4) lots experienced around (b) (4) or less time out of the incubator. Given the necessity of providing the intended recipient the product once RATGAM treatment has been performed, it will be important to assure that the intended shelf life is sufficient. Your package insert should advise that expired product lots should not be used.

The proposed expiration should be evaluated in the new PV study to support the final DP expiration.

Meeting Discussion for Applicant Question 16:

There was no discussion of this question during the meeting.

Applicant Question 17:

- *Does the Agency agree that the process validation studies outlined in [Table 10](#) are sufficient to demonstrate manufacturing and product consistency for all elements?*
- *Process validation will be performed on (b) (4) lots that may be used to treat patients under the IND. Does the Agency agree that, in the unlikely event that all slices from a given lot must be administered to the patient in order to achieve the minimum dose, and thus no slice from that lot can be returned to the (b) (4) facility for use in histology testing, this lot may be considered invalid and an additional lot could be manufactured for PV?*
- *Enzyvant also seeks to discuss the possibility of submitting the process validation report on a rolling review basis, as described in [Section 11.14](#).*

FDA Preliminary Meeting Response to Applicant Question 17:

The design of the new process validation study is an improvement over the previous study; however, we still have concerns. Your proposed process validation study is not adequate because it does not sufficiently examine sources of variability and provide reasonable assurance that in-process and final product controls you have in place help assure manufacturing consistency, as described in the FDA process validation guidance and ICH Q7, and as communicated in the August 7, 2018 Type C meeting.

Our understanding is that you propose to generate (b) (4) additional (or a minimum of (b) (4) clinical lots/PPQ lots that adhere to the most recent versions of step and hold times and meet all in-process and lot release criteria. The release specifications will include the revised histology acceptance criteria. Because these lots will be used for treating patients, the time of harvest will depend upon patient scheduling, unless no patient is scheduled, whereby the PPQ lot will be released between Day 14-21. The tests will also be conducted after the revised material and personnel flow procedures have been implemented, and after the final container and secondary container transport study have been completed.

For the purpose of this study (but not for future commercial lots) you will also verify that the required Day (b) (4) filter coverage of (b) (4) is achieved. You will also include a calculation of yield based on the number of tissue slices manufactured and released, which will be calculated daily for each lot.

Your design is an improvement over the study reported in the original submission because all (b) (4) lots will be processed generally the same way, in accordance with your manufacturing protocols for clinical production. Despite the fact that the new validation study will encompass the new changes to your facility and protocols, and will include evaluation of filter coverage and yield, we still find the study insufficient for the following reasons:

1. The design does not include important assessments, including the following:

- a. There is no examination of slice thickness produced using the tissue slicer. While we appreciate it is your opinion that slice thickness is not important clinically, you have not provided any clinical data on patients who received only thin slices. Most clinical lots were composed of a mixture of thick and thin slices, with the majority being thick. A small number of patients received only thick slices. It is not clear the degree to which thin slices contribute to clinical outcome, and it does not appear that biopsy data can inform on whether the biopsy is from a thick or thin slice. We acknowledge that no validated assay is currently in place for determining slice thickness, and that you stopped assessing thickness once the manufacturing process was transferred to the (b) (4) facility. However, the slicing of tissue is a critical manufacturing step, and you should verify that this unit operation is suitable for the intended purpose and consistent with data generated to support safety and efficacy. For the purpose of the process validation study it would be acceptable to use whatever assessment was in place in the (b) (4) .
- b. You have established a (b) (4) minimum acceptance criterion for further processing into tissue slices. It is unclear how this value was determined. It is also unclear how consistent that determination is given the tissue is (b) (4) before trimming and some tissues have (b) (4) associated with them, such as after being held (b) (4) in medium versus supplied fresh in a (b) (4) container. There is also little correlation between the amount of tissue supplied by the operating room and the number of slices generated. We recommend that you monitor the number of slices generated (b) (4) of tissue. We further recommend that you set a target number of slices to culture in addition to the minimum and maximum number of slices required. For the safety and efficacy lots the average number of slices transplanted was (b) (4).
- c. According to research publications by (b) (4), the upper limit for days in culture was set at 3 weeks because beyond this time (b) (4) was a potential concern. Please provide some measurement or general assessment of (b) (4) in your study to demonstrate that up to 21 days of culture is appropriate. You may wish to consider incubating additional slices for longer than 21 days for comparison purposes.
- d. The ability to remove slices from the filters for surgery without damage is not assessed. During discussions held during review of the original submission, and on facility inspection, it became apparent that the process of removing of the slices may be more complicated than described. The product has variability not only in the thickness of the slices, but substantial variation in size. To our knowledge no histological evaluation of slice size and thickness and potential damage to the slices during the process of removal from the filter has been examined. If no previous assessment has been performed, we recommend that you remove the smallest and thinnest slices from the filters and compare the quality to larger, thicker slices from the same

lot.

- e. We agree it is important to calculate yield on the cultured slices, but as you indicate, you have no reference because this data has not previously been captured. You have not indicated how you will use the collected data in your assessment of manufacturing consistency and no predefined criteria were described.
2. Because the PPQ lots are intended to be used for patient treatment, there is no guarantee that these lots will represent worst case and include the maximum length of time in culture or be held for the full hold period, including (b) (4) hold at (b) (4). While worst case conditions are not necessarily a requirement for process validation, limited data are available. Clinical data on individual processing steps or hold times does exist and is supportive of individual conditions, but clinical data does not exist for the maximum processing conditions at all steps in combination. Limited information also exists demonstrating manufacturing consistency. Including worst case conditions for your PPQ lots would help address these concerns. However, we do not recommend testing the totality of worst-case conditions
3. It is not clear that PPQ lots in your proposed study would allow sacrificing an additional slice at Day 12, as was part of the previous study, and there is no guarantee that any one lot will be harvested at either the minimum 12 days or maximum 21 days as allowed by protocol. Your PPQ study should evaluate manufacturing consistency across the full allowable culture period.

We appreciate your desire to avoid delays in patient treatment for this very rare, and severe disease, and acknowledge the scarcity of the source material. We understand that dedicating source material for three consecutive process validation lots could represent a significant delay in patient treatment until new source material is available. We offer the following suggestions:

1. Though FDA guidance recommends conducting process validation on (b) (4) lots, this therapy and patient population presents special challenges, largely due to serious limitations on source material availability. For this reason, it would be acceptable to not use consecutive lots, though the sequence of lots used should be documented and justified, and you should indicate how the selection of source material for PPQ versus clinical lots does not induce bias.
2. You may wish to consider using a (b) (4) manufacturing approach, where (b) (4) lots of source material would be used to produce (b) (4) PPQ lots (b) (4). This would reduce the number of source material collections to (b) (4) versus (b) (4). Following slicing, slices should be (b) (4).

All lots would be held to the maximum at each process step and held (b) (4). Each (b) (4) lot would be tested according to all procedures and include additional testing as discussed above. The (b) (4) lots would not have to meet the minimum (b) (4) slice specification, as

long as all the specified testing can be performed. Should the source material be sufficient to allow each (b) (4) lot to meet your minimum (b) (4) slice acceptance criteria for commercial production, then the same procedures for commercial production should also be followed. Since the lots would be (b) (4) you should perform a secondary comparison of each (b) (4) lot for consistency, in addition to an evaluation of consistency across all (b) (4) lots.

3. If you do intend to use PV lots to treat patients, please be sure to submit an amendment with the final PV protocol to IND 9836. We recommend that you await FDA feedback before proceeding with the process validation study, unless agreement has previously been reached with the Agency on the plan.

Meeting Discussion for Applicant Question 17:

Enzyvant indicated they intend to follow the option FDA provided on using (b) (4) lot manufacturing for the process validation study. As recommended, they will produce (b) (4) new lots, each (b) (4) lots, to generate a total of (b) (4) lots. (b) (4)

In addition to in-process and final product testing, the process validation (PV) study will also incorporate estimation of slice thickness (b) (4) technique); (b) (4) will be examined histologically, including culturing additional slices (b) (4) both thick and thin slices cultured for up to 21 days will be examined, and, to address concerns about possible damage to slices during removal from the filter, slice quality will be assessed after removal; and maximum hold times will be used for PV lots, including incoming thymus transport, final drug product delivery, and (b) (4) hold of incoming thymus at (b) (4). PV lots will not be used to treat patients.

FDA sought clarity on how Enzyvant will measure manufacturing yield. Enzyvant indicated yield would be a measure of the number of slices at the time of culture initiation and at the end of culture, which is expected to be (b) (4). Any deviation from (b) (4) yield will be investigated. FDA recommended a different strategy because such data would not be informative since the tissue slices adhere tightly. Enzyvant confirmed they have never lost a slice during manufacturing. FDA clarified that yield calculations as part of process validation would be directed at demonstrating that similar numbers of slices are generated (b) (4) of source material tissue. FDA's evaluation of Enzyvant's manufacturing history indicated a low correlation between the amount of starting material and the number of slices generated to initiate the cultures and the final dose obtained. During pre-license inspection, it was also noted that (b) (4) of the tissue occurs (b) (4), and that (b) (4) tissue can lead to discrepancies. Enzyvant clarified that the tissue is always (b) (4) hold in culture medium, but that the amount of (b) (4) the tissue could be a factor. FDA recommended they capture (b) (4) images of the entire slice of each PV slice including on Day (b) (4) so that FDA has a better idea of what the slices look like. During pre-BLA discussions they had provided such images as a time course and that was very helpful to understanding the consistency of their manufacturing process. Enzyvant agreed to capture such images but noted that defining the slice border can be complicated by the presence of (b) (4).

Enzyvant indicated the study is not intended to test worst-case conditions of daily media changes (minimum and maximum time between changes) or the maximum number of lots of thymus organ medium (TOM) used per lot of drug product, which had previously been up to (b) (4) different lots. Enzyvant proposed testing using (b) (4) different lots during manufacturing, as is the typical situation. FDA indicated this would be acceptable but recommended an evaluation of performance of different batches of culture medium or fetal bovine serum (FBS). FBS is well-known to vary considerably lot-to-lot for some cell cultures, and since Enzyvant does not conduct performance qualification on FBS or TOM culture medium prior to use for clinical manufacturing, they may wish to perform small scale pilot studies to evaluate this variable.

Enzyvant indicated they would follow flexibility offered by the FDA that PV lots will not have to adhere to the (b) (4)-slice minimum for clinical lots but will have to have sufficient numbers of slices to complete all tests. Since the scale may be below the minimum that FDA recommended, Enzyvant should provide some bridging data in their resubmission. For example, they could compare results obtained for the small-scale PV study lots with previous lots produced at maximum scale.

Enzyvant stated it was not their intention to submit a revised PV protocol for FDA's review prior to conducting the studies and asked if that approach was acceptable. Submission of a protocol for FDA's review is at Enzyvant's discretion as there is no requirement. FDA informed Enzyvant that if they do submit a copy for review, FDA will try to provide a response shortly, but due to workload cannot promise feedback by a specific date. FDA advised that following the basic outline of the (b) (4) manufacturing approach offered is acceptable; however, in their BLA resubmission they should justify why this strategy adequately encompasses the specific elements of their manufacturing process and satisfies the goals of process validation. The PV study should demonstrate that each unit operation and the manufacturing process as a whole are adequately controlled to ensure manufacturing consistency.

Applicant Question 18: Does the Agency agree that, given the information described above, successfully completing a DP transport study as described above and including the reports in the BLA resubmission will demonstrate that the final DP container adequately maintains a sterile environment and will address this CRL deficiency?

FDA Preliminary Meeting Response to Applicant Question 18:

Your proposal for the new study appears to be reasonable. The final decision of its adequacy will be reached during the BLA review.

Meeting Discussion for Applicant Question 18:

There was no discussion of this question during the meeting.

Applicant Question 19: Does the Agency agree that the controls and sterility testing described above suitably assure sterility of the (b) (4) container?

FDA Preliminary Meeting Response to Applicant Question 19:

Your proposed sterility testing approach for incoming lots appears to be acceptable if supported by a properly validated sterilization cycle. We reviewed the sterility validation report when it was initially submitted to the BLA and you did not provide sufficient

additional information in the meeting package to change our conclusions about the validity of the sterilization study. Specifically,

1. It is still not clear whether the (b) (4) container is representative of (b) (4) with respect to bioburden and sterilization challenges. Please clarify whether both containers are manufactured using the same materials and process and have similar dimensions (i.e. opening diameter) and secondary packaging configuration.

2. The (b) (4) study results provided in the meeting package cannot be interpreted, as neither narrative nor (b) (4) placement diagram was included. Furthermore, (b) (4) was performed almost 9 years prior to the sterilization validation. This could be acceptable if you can confirm that (b) (4) was performed similarly in both studies (e.g., same package orientation and number of (b) (4) E-beam was regularly maintained, and packaging of the test item did not change since 2008. Please note that for the validation study to be acceptable, (b) (4) had to be placed in the worst-case location, as determined during the (b) (4).

Meeting Discussion for Applicant Question 19:

There was no discussion of this question during the meeting.

Applicant Question 20:

- *Does the Agency agree that successfully repeating the thermal quality study, which will be designed similarly to the original shipping temperature validation study, with the new drug product shipping container will resolve this deficiency?*
- *Does the Agency agree that a standard distribution simulation test does not need to be conducted on the drug product shipping configuration, since the culture dishes must be handled with care and cannot withstand shaking or dropping during transport?*

FDA Response to Question 20:

Your proposal for thermal quality study is acceptable. Please note that for the study results to be valid, you should meet the acceptance criteria for the temperature outside of the cooler throughout the entire duration of your study.

We agree that a standard distribution test does not need to be conducted. The purpose of the proposed modified handling study is not clear. If you decide to proceed with this study, please see our advice regarding leak detection below (response to Q. 21).

Meeting Discussion for Applicant Question 20:

There was no discussion of this question during the meeting.

Applicant Question 21: *Does the Agency agree that the cleaning and packing procedures, as well as the spill procedures, are appropriate and address this cited deficiency?*

FDA Response to Question 21:

We cannot comment on the appropriateness of your cleaning and packing procedures at this time, as it is dependent on the outcomes of your transport validation and (b) (4) cleaning validation studies. For your study:

1. Please ensure you validate the maximum clean hold time of the secondary container unless you intend to use it immediately after cleaning.
2. We recommend you carefully consider and train operators on what constitutes a spill/leakage and how it is detected upon delivery of the product to the operating room (OR), as not all leakage might be immediately apparent during the visual inspection of a closed tissue culture dish. Given that the tissue culture dishes are stacked during transport to the OR, please also consider impact of a leaking top dish on the dishes underneath it.

Meeting Discussion for Applicant Question 21:

There was no discussion of this question during the meeting.

Applicant Question 22:

Does the Agency agree that conducting the proposed (b) (4) system qualification and implementing the described routine (b) (4) sampling, and fully describing this in the BLA resubmission, will adequately address this cited deficiency?

FDA Response to Question 22:

Requalification of the (b) (4) system and implementation of a consistent routine (b) (4) sampling is a step in the right direction. We cannot comment on whether it can adequately address the deficiency as that would depend on details of the study design, acceptance criteria and testing used, and the outcomes of the study. Please also include the following information when responding to this deficiency,

- A justification of sampling duration during the qualification of the (b) (4) system and for routine sampling locations, unless all POU's are sampled (b) (4).
- A detailed description and predetermined acceptance criteria for all testing performed, including routine monitoring testing.

Meeting Discussion for Applicant Question 22:

There was no discussion of this question during the meeting.

Applicant Question 23: *Does the Agency agree that implementation of the planned changes may adequately address this deficiency?*

FDA Response to Question 23:

The proposed changes are an improvement over the current situation as they (b) (4)

However, we cannot comment at this time whether this will address the deficiency, as that would depend, among other things, on the outcome of the new EMPQ. Additionally, we have the following comments:

1. You should ensure sufficient pressure differentials are being implemented between differently classified areas in the modified facility. The pressure differentials should be continuously monitored, recorded, and connected to an alarm.

2. All active pass-throughs should be classified and qualified to meet the classification; recovery time should be determined and used for timing of the interlocking doors, as appropriate.
3. Your traffic light system setup is not clear. When responding to the CRL, please provide room numbers and doors where the system is installed and what triggers the on/off signal. Please note that your system should ensure that personnel exits the dirty corridor before additional personnel enters it from the manufacturing rooms.
4. From the product perspective, it is acceptable for manufacturing personnel to exit the facility out of Gown-Out and return to Changing Room for changing into street clothes. The personnel must change into new set of scrubs if they are re-entering the Receiving/Supply Room.
5. It is acceptable to include additional supplies in your toolboxes. However, given that your toolboxes are process specific, please clarify how you plan to use additional supplies (e.g. repackaging within the facility, use the same toolbox for multiple lots, etc.) and to maintain tracking and tracing of materials.
6. If you allow personnel to remain in hallways (“hallway monitors”), any associated activities as well as the total occupancy of the corridors should be validated during EMPQ.

Meeting Discussion for Applicant Question 23:

There was no discussion of this question during the meeting.

Applicant Question 24:

- *Does the Agency agree that providing the process validation reports to the FDA following submission of the other components of the BLA resubmission, as part of a rolling review, can be acceptable for the RETHYMIC BLA resubmission?*
- *Given the extent of ongoing work to address the FDA concerns cited in the CRL, Enzyvant would like to continue to partner and work closely with the FDA leading up to the BLA resubmission. Is the Agency amenable to additional meetings and correspondence with Enzyvant prior to the BLA resubmission?*

FDA Response to Question 24:

We appreciate your interest in trying to expedite the resubmission of your BLA, but we do not agree with your approach of providing process validation data as a rolling submission. We do not feel that this type and quantity of critical manufacturing data falls under the category of special circumstances where additional CMC information could be provided after Module 3 is submitted. Examples of CMC information that could potentially be provided after BLA submission could include amending stability data provided in the submission with longer stability time points in order to provide a more complete stability report or providing a revised batch analysis based on additional product lots completed after submission of Module 3. These final data sets could be evaluated separately from the main submission and would not be expected to significantly impact other sections of the BLA review. Process validation is central

to demonstrating manufacturing consistency, in providing support that the manufacturing process is in an adequate state of control, and that assays and procedures are suitable for the intended purpose. We feel it would be unproductive to submit these studies at a later date. Further, considering the unpredictable nature of when tissue will be available, whether it will meet the minimum processing criteria, and since it is your intention to transplant into patients, whether it will meet donor eligibility, it is not clear when we could expect results from all process validation lots to be available. Once Module 3 is submitted the PDUFA clock would start. Providing critical data late in the review cycle will only complicate BLA review.

You have requested feedback on plans to respond to BLA CRL items and request additional advice and opportunities related to the resubmission of your BLA. You indicate that based on urgent medical need for this patient population, you wish to resubmit as soon as possible. If you feel you need additional feedback on specific topics not covered in our responses, options exist to obtain additional advice:

- You may provide copies of your study designs in advance of execution as product correspondence amendments to the BLA. Please note that due to workload issues we cannot guarantee a quick review, and suggest you submit well in advance of when you plan to execute.
- You may request an additional meeting(s). We recommend that future meetings be requested as Type B meetings and to make the best use of such meetings that you cover areas across multiple disciplines, including any potential clinical questions. You should plan on including updated safety and efficacy data in Module 5, including data on product lots manufactured in the (b) (4) facility. Questions about product labeling would be best handled once the BLA is resubmitted.

We offer the following general advice for your resubmission to improve the quality of your submission and avoid numerous information requests during the review process:

- Your BLA resubmission should be as complete as possible. Information requests and Mid and Late Cycle meetings are not an efficient means to compensate for information that should be in the original submission. Responses to information requests should be to provide a finer level of detail and provide additional clarity.
- Please be sure any information included in your submission is accurate and up to date, and that all referenced documents have been harmonized with the same information. Information in Modules 3 and 5 should be consistent and support each other.

Meeting Discussion for Applicant Question 24:

There was no discussion of this question during the meeting.