

Our STN: BL 125685/0

COMPLETE RESPONSE
DECEMBER 4, 2019

Enzyvant Therapeutics Inc.
Attention: Kevin Healy, PhD
300 Morris Street, 7th Floor
Durham, NC 27701

Dear Dr. Healy:

Please refer to your rolling Biologics License Application (BLA) submitted July 6, 2018, received April 5, 2019, for Allogeneic Processed Thymus Tissue manufactured at your (b) (4) location and submitted under section 351(a) of the Public Health Service Act.

We have completed our review of all the submissions you have made relating to this BLA with the exception of the information in the amendments submitted and received November 15, 2019 and November 26, 2019. After our complete review, we have concluded that we cannot grant final approval because of the deficiencies outlined below.

Chemistry Manufacturing, and Controls

1. Outstanding issues identified during the pre-license inspection (PLI) at your contract manufacturing facility conducted (b) (4), as detailed in Form FDA 483, have yet to be resolved. Please submit documentation that demonstrates that all outstanding inspectional issues identified during the PLI have been resolved.
2. The proposed sampling and testing strategy for your histology-based potency assay is not acceptable because it neither fully supports the need for making an informed decision on initiating treatment of the patient with rabbit anti-thymocyte globulin (RATGAM) nor provides an adequate assessment of the product the patient will receive. Please address the following concerns:
 - a. (b) (4) testing: We agree that the risk RATGAM treatment presents to the intended RETHYMIC patient makes it important to have confidence that the intended product lot is consistent with lot release at the time of interim testing. Your proposed strategy appears to be inconsistent with this goal. Please revise your strategy to take into account the following:

- i. Even if histology results from early sampling, as proposed, meet your acceptance criterion, the product lot could fail for other reasons. Results from donor qualification should also be known but are not available until Day 12. Further, in-process testing for (b) (4) should be implemented.
 - ii. If there are delays in the scheduling of the transplant that extend beyond the (b) (4) window, you do not have a plan in place once this window is exceeded, such as discarding the product lot or retesting.
 - iii. You indicate that if histology testing spans (b) (4), then more than (b) (4) may be needed to obtain results. In such a case, if RATGAM treatment involves 3 daily doses followed by 2 days prior to transplant, it does not appear the product lot could be released within 8 days.
 - b. Testing of the final product: You failed to demonstrate that testing (b) (4) by (b) (4) is reasonably representative of the drug product (DP). Please implement testing by histology on another slice taken from the drug substance as close as is feasible for product release. Please propose a window of histology sampling for DP release.
3. Please address the following deficiencies related to the histology assay used to assess product safety and quality including identity, potency, and purity:
 - a. You provided the Histology Training Guide that serves as a training manual for pathologists performing evaluation of RETHYMIC. However, there are no Standard Operating Procedures (SOPs) for the procedures performed by the pathologists. Written procedures are required for both manufacture and process controls designed to ensure that the DPs have appropriate levels of identity, strength, quality, and purity (21 CFR 211.100). Please provide an SOP for the histological evaluation.
 - b. The histology assay performed during method validation implemented (b) (4) criteria. The use of (b) (4) criteria is a more rigorous reflection of the depth of analysis performed by the pathologists than the (b) (4) criteria you proposed for release testing and provides further assurance of product quality. Furthermore, your batch records reported both (b) (4) and (b) (4) histology results, including reporting a percentage of (b) (4). Thus, please revise your histology acceptance criteria to include a (b) (4) measure for assessment of (b) (4).
 - c. Data from twenty-nine subjects treated under IND used to support the safety and efficacy of RETHYMIC documented low naïve T cell counts

during the first 2 years post-transplant. You have not adequately excluded the possibility that these patients received lower quality lots. You indicated that histology testing met the release criteria for these product lots but did not provide examples of histology images from patients who had positive or negative clinical outcomes. In order to establish a basis by which (b) (4) histology results can be evaluated, please perform a retrospective (b) (4) histological analysis of product lots used to support clinical safety and efficacy, including new product lots produced in the (b) (4) facility. Please discuss how the retrospective analysis supports the setting of (b) (4) histological criteria. Please also include examples of the evaluated histology images in your BLA resubmission.

4. Support for a thymus source material hold time of (b) (4) is insufficient:
 - a. Manufacturing instructions for clinical lots produced under IND in the (b) (4) state the tissue was to be immediately processed or stored (b) (4). Thus, the data supporting clinical safety and efficacy appear to be based on source material handled differently than what you propose in the BLA. You did not provide any additional clinical data based on source material held for (b) (4).
 - b. Your process validation (PV) study intended to support the (b) (4) hold time was based on (b) (4) histology results from a single tissue slice from a single lot (b) (4) held at room temperature in a (b) (4) specimen cup. The study is insufficient to determine whether the (b) (4) profile for overall quality on the Day 21 slice represents adequate product quality, and whether the assay is sufficiently sensitive to support stability of the tissue under these storage conditions. No (b) (4) assay was included in your analysis.
 - c. You propose a maximum hold time of (b) (4) in the operating room (OR) from notification of thymus harvest up to time of pick-up; however, PV lot (b) (4) was only exposed to a hold time of (b) (4). Other (b) (4) lots have been held for as long as (b) (4), but those were not exposed to the same conditions as lot (b) (4). You propose a total hold time of (b) (4) from the time of notification of tissue availability, but this does not factor in the full (b) (4) of OR hold time.

To support your proposed full hold time, you will need to either 1) provide historical clinical data from patients treated with source material held for (b) (4) that covers all intended maximum step times, 2) conduct a stability study using a (b) (4) stability-indicating assay on multiple lots tested using multiple slices, or 3) establish a shorter expiry based on historical clinical data of safety and efficacy.

5. You propose an expiry of (b) (4) for the Drug Product (total time outside incubator until end of surgery) based on one clinical lot manufactured in 2018 that experienced a hold time of (b) (4) minutes. However, these data do not support the proposed hold time because that lot was formulated and transported in the (b) (4) container, not the (b) (4) final product container, and therefore does not represent the commercial process. Clinical data provided on product lots packaged in the (b) (4) tissue culture dish that were manufactured and transported from the (b) (4) and the (b) (4) facility were held for less than (b) (4) between product formulation and administration. Please establish an expiry based on relevant clinical data using the proposed formulation and (b) (4) final product container or provide additional stability data using a (b) (4) stability-indicating assay.
6. The PV study does not adequately demonstrate manufacturing and product consistency for all elements. A successful PV study should demonstrate that each unit operation is performing as intended, and manufacturing is consistent lot-to-lot. However, this was not fully demonstrated in Process Validation CT2-2017-013-P. Please perform an additional study to address the following concerns:
 - a. Unlike (b) (4) staining performed for the purposes of identity, which had successfully demonstrated substantial reduction in donor thymocyte levels by Day (b) (4) and the presence of key hallmarks of thymus tissue at all stages, the same methods applied to potency and overall tissue quality are not conclusive for the following reasons:
 - i. Results of this study and other data provided in the submission show wide variation in the phenotype of tissue slices and the expression pattern of (b) (4) within different regions of the same slice, different slices, different lots, and different culture times.
 - ii. Though all lots and time points met release criteria, the criteria are broad, raising concerns about the sensitivity of the assay.
 - iii. The impact on tissue slice quality is difficult to assess because data on the tissue received by subjects treated under IND was not provided (no retrospective comparison was made of product lots received by subjects who had either a positive outcome, negative outcome, or a reduced/delayed naïve T cell development).
 - iv. (b) (4) staining profile in the PV lots appears to change to a greater degree over the course of 21 days compared to other time course examples provided to date. It is unclear if this is related to

differences between lots or to longer step and holding times included in the PV study intended to represent worst case.

- b. The design of the study is complicated by the range of variables included in the study. No two lots were treated the same way, and no one lot was exposed to the maximum conditions at all stages. While we appreciate your efforts to cover the range of conditions the lots would be exposed to for commercial manufacturing, PV is typically performed after critical process parameters for all manufacturing steps have been established. In general, for a PV study, a minimum of (b) (4) lots should be manufactured under the same conditions. The information provided in this study is typically reported under Sections 3.2.S.2.6 (Manufacturing Process Development) or 3.2.S.2.4 (Control of Drug Substance). You also considered a process step to be validated based on the outcome from (b) (4). Further, the study was designed to use (b) (4) culture medium per lot, yet most clinical lots used (b) (4) lots, and you report that up to (b) (4) lots of culture medium were used for some clinical lots. Testing of your manufacturing process should represent conditions typically used.
- c. Unit operations:
- i. The extension of the culture medium exchange time to (b) (4) is not adequately supported, since (b) (4) intervals were not tested on (b) (4) medium exchanges for cultures beyond Day (b) (4). The study is also limited by the fact that only one product lot was exposed to these conditions, and there was no comparison made to elucidate the effects of these conditions on thick versus thin or small versus large slices.
 - ii. At the initiation of culture, the (b) (4) filters must be covered with (b) (4) of tissue, but no (b) (4) method was used to verify filter coverage.
 - iii. The clinical data set indicates that about (b) (4) of slices produced for clinical lots with the tissue slicer are thick and (b) (4) are thin, though the proportion varies by product lot. Slice surface area varies greatly within a lot and between lots. No evaluation of the consistency of slice thickness or size was included in the study. Since it is unclear whether slice thickness or size has a meaningful impact on clinical outcome, the commercial process should be better controlled to maintain consistency in the properties of clinical lots.
 - iv. No calculation of yield was performed, and no comparison was made with clinical lot production under IND.

7. Transport study (b) (4)-2019-050-A failed to demonstrate microbial protection of DP during packaging, transportation to the OR, and hold in the OR in the (b) (4) culture dish and (b) (4) secondary container. If you intend to proceed with commercialization of the (b) (4) final DP container, please investigate the media growth promotion failures and take appropriate corrective actions prior to conducting a new study demonstrating that the final DP container adequately maintains a sterile environment. Please submit the summary reports.
8. You failed to assure sterility of direct product contact materials. Specifically, (b) (4) validation of the (b) (4) container used for source material transport and (b) (4) storage was deficient. The study was performed on a different container, and (b) (4) was not performed. Please provide the summary report for sterilization validation of the (b) (4) container.
9. Adopting the (b) (4) culture dish as your primary DP container changed your DP packaging and configuration of the shipping container used for DP transport to the OR. Therefore, the validation of this shipping container to maintain the appropriate temperature is no longer valid. Please revalidate and provide the summary report.
10. Due to the nature of your primary DP container, the environment inside your secondary (b) (4) container becomes more critical to ensure microbial protection of the product. We recommend cleaning and/or sterilization validation of the secondary container and packing of the (b) (4) container in the ISO (b) (4) environment. Additionally, please implement and provide procedures and lot disposition for spill incidents in transport.
11. Regarding your (b) (4) system:
 - a. Qualification of your (b) (4) system is deficient in scope and duration. Specifically, it did not include monitoring of (b) (4) quality over a period of time, and only a limited number of locations were sampled. (b) (4) sampling did not demonstrate that (b) (4) is within ISO (b) (4) acceptance limits.
 - b. Your strategy and schedule for routine (b) (4) sampling is unclear, as not all testing is performed quarterly, and locations vary for different dates and types of tests. The sampling procedure description is inconsistent (e.g., use of (b) (4) and vague about (b) (4) use during sampling, which could interfere with (b) (4) testing.

Please provide information and/or data to address these issues.

12. The personnel flows at your multi-product facility create an increased risk of product contamination and cross-contamination. Specifically,
- a. You allow (b) (4) [REDACTED] of your facility. This allows simultaneous presence of personnel working on different products in (b) (4) [REDACTED].
 - b. Additionally, personnel enter Gown-In Room (b) (4) [REDACTED] and exit Gown-Out Room (b) (4) [REDACTED] of the facility through the same Receiving/Supply Room (b) (4) [REDACTED]. This allows simultaneous presence of personnel entering and exiting the manufacturing areas in Room (b) (4) [REDACTED].

Please provide a description of procedural and/or engineering controls in place to ensure appropriate personnel flows, to prevent exceeding the maximum number of allowed personnel in Rooms (b) (4) [REDACTED], and to mitigate risk of product contamination and cross-contamination due to personnel flows described above.

13. We reserve comment on the proposed labeling until the application is otherwise acceptable. We may have comments when we see the proposed final labeling.

Within one year after the date of this letter, you are required to resubmit or withdraw the application (21 CFR 601.3(b)). If you do not take one of these actions, we may consider your lack of response a request to withdraw the application under 21 CFR 601.3(c). You may also request an extension of time in which to resubmit the application. A resubmission must fully address all the deficiencies listed. A partial response to this letter will not be processed as a resubmission and will not start a new review cycle.

You may request a meeting or teleconference with us to discuss the steps necessary for approval.

Please submit your meeting request as described in the guidance for industry *Formal Meetings Between the FDA and Sponsors or Applicants of PDUFA Products* at <https://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM590547.pdf>, and CBER's SOPP 8101.1 *Scheduling and Conduct of Regulatory Review Meetings with Sponsors and Applicants* at <http://www.fda.gov/BiologicsBloodVaccines/GuidanceComplianceRegulatoryInformation/ProceduresSOPPs/ucm079448.htm>.

If you have any questions regarding the above, please contact the Regulatory Project Manager, Jean Gildner, at (240) 402-8296 and Adriane Fisher, at (301) 796-9691.

Sincerely,

Wilson W. Bryan, MD
Director
Office of Tissues and Advanced Therapies
Center for Biologics Evaluation and Research