

CBER CMC BLA Review Memorandum

BLA STN 125685

Allogeneic processed thymus tissue-agdc

(RVT-802, RETHYMIC)

Enzyvant Therapeutics GmbH

Reviewers

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1. **BLA#:** STN 125685

2. **APPLICANT NAME AND LICENSE NUMBER**

Enzyvant Therapeutics GmbH

3. **PRODUCT NAME/PRODUCT TYPE**

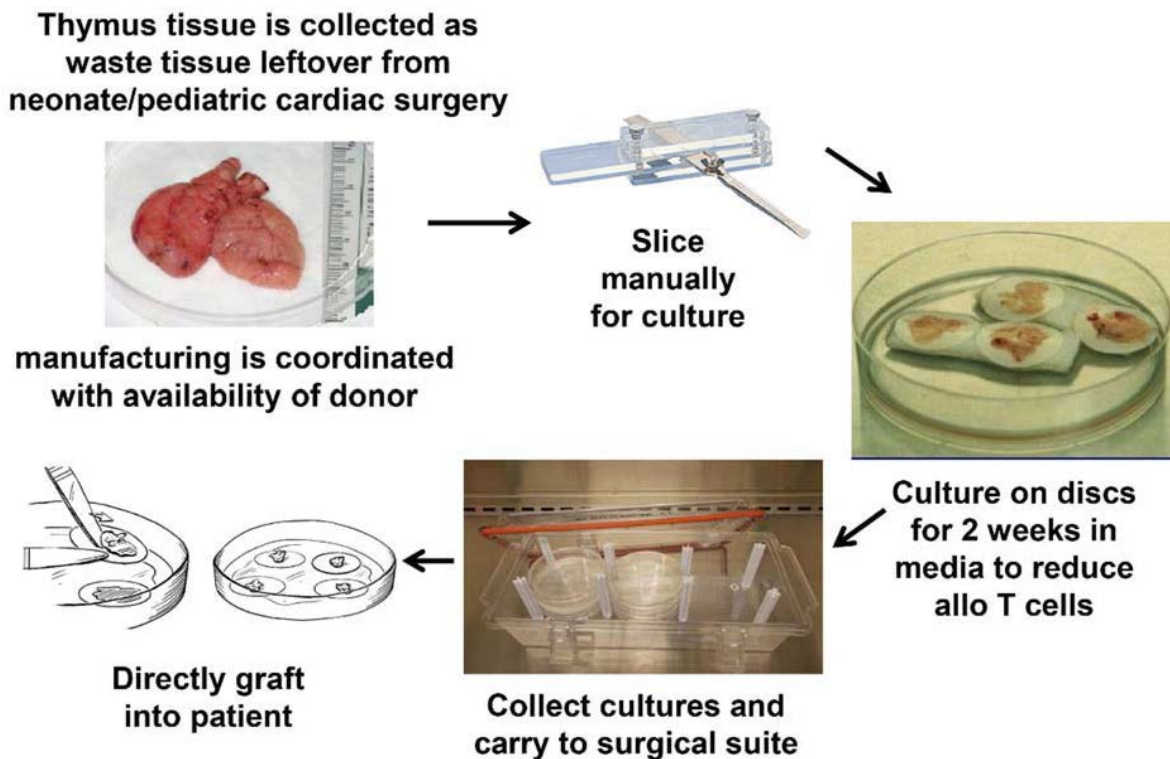
Non-Proprietary/Proper/USAN: Allogeneic processed thymus tissue-agdc

Proprietary Name: RETHYMIC

4. **GENERAL DESCRIPTION OF THE FINAL PRODUCT**

The product is neonatal allogeneic, unrelated thymus tissue slices cultured in (b) (4) medium supplemented with (b) (4) fetal bovine serum (FBS) for up to 21 days intended as a treatment for congenital athymia. (b) (4) medium is applied (b) (4) to culture by (b) (4) onto the thymus slices (b) (4). The slices are surgically implanted one at a time into thigh muscle of the patient. A “slice” is defined as the total amount of material covering a (b) (4) filter. The slice generally covers about a quarter of the surface area of the filter. Filters are suspended on a piece of surgical gel foam above the culture medium. A slice can be the sum of multiple slices placed on the same filter at the time of culture. Slice thickness varies from approximately (b) (4). A product lot is intended for a single patient and can be composed of up to (b) (4) filters, with up to 4 filters per (b) (4) culture dish (final container closure). We have summarized the overall manufacturing process in Figure 1.

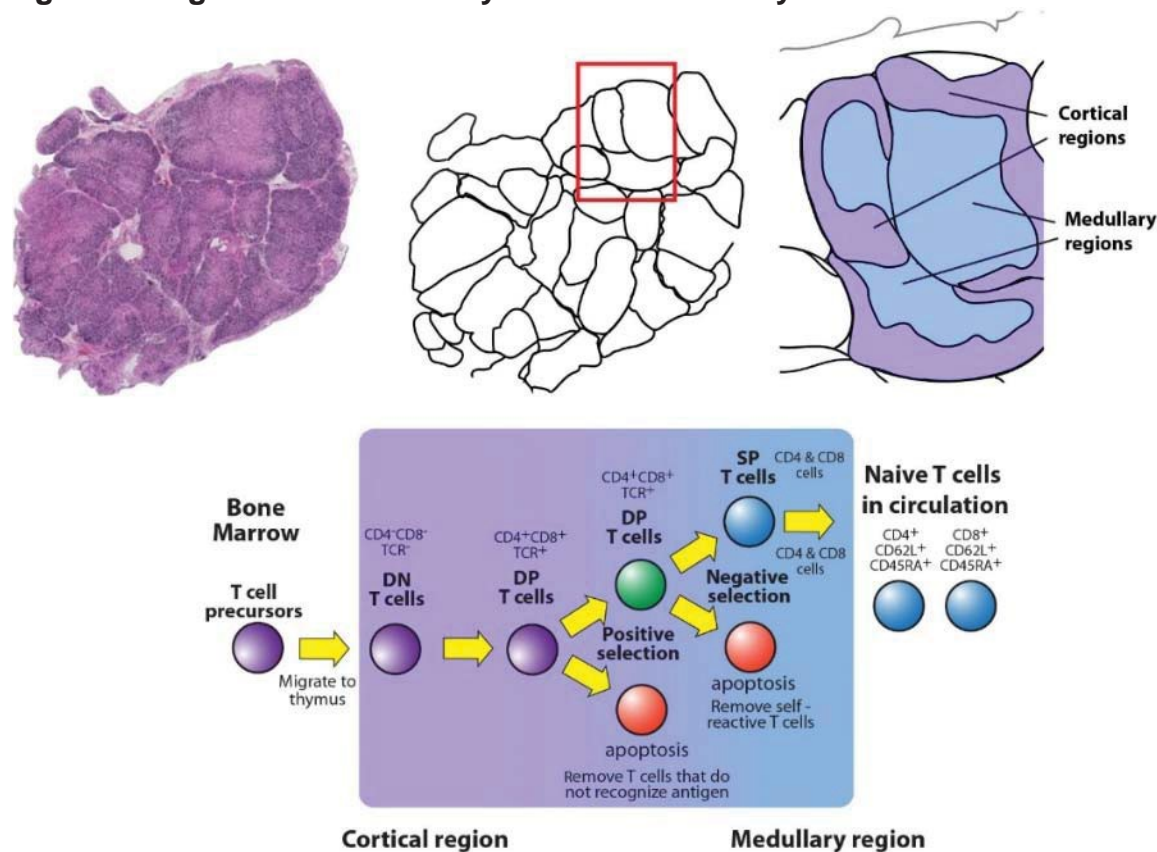
Figure 1: Manufacturing overview.



The active ingredient is considered the entire slice. Dose is based on the total surface area of all slices, divided by the body surface area of the patient (b) (4) body surface area formula). The allowable dose range is (b) (4) – 22,000 mm² slice area/m² body surface area and is based on the dose range experienced. The product is formulated in the same culture medium as for drug substance generation and is composed of (b) (4), and (b) (4) FBS. The final container closure is the same set of (b) (4) culture dishes used for culturing, but the (b) (4) of media used during culture is replaced with 5 mL of medium as an excipient. with the only difference being 5 mL of culture medium is used as excipient instead of (b) (4) used during culture. The dishes are transported in a clear acrylic secondary container (b) (4) container) placed inside an insulated (b) (4) cooler with (b) (4). The proposed shelf life is (b) (4), which covers packaging, transport, and surgical times. The product is transported by (b) (4) facility members who participate in the surgical procedure by handling the dishes for the surgical team and record the number of slices used. Any remaining slices that were not transplanted are transported from the surgical suite back to the (b) (4) facility, the transplanted dose is calculated at the (b) (4) facility, and any remaining product that was not transplanted is discarded.

Thymus is composed of a cluster of lobules that represent repeating subregions of tissue. As depicted in the illustration we generated below (Figure 2), each lobule contains cortical and medullary regions. These regions have specific functions in T cell maturation and education.

Figure 2: Organization of the thymus and role of thymus in T cell maturation.



Immature bone marrow T cells are recruited to the thymus via chemokines. Once in the thymus they move from a cortical-medullary junction into the cortical portion of a lobule. In the cortical region they encounter multiple cell types. Of special importance are cortical thymic epithelial cells that act as antigen presenting cells. After undergoing maturation, the T cells undergo positive selection and T cells that do not recognize antigen in the context of MHC are eliminated. The T cells then move to the medullary region where they undergo negative selection to remove self-reactive T cells. The medullary region is also important in Treg production. Negative selection by RVT-802 in transplanted patients must also include tolerance to donor antigens.

The organization of the thymus and distribution of cortical and medullary thymic epithelial cells is important. A slice of thymus tissue cuts through multiple lobules, and the slices are thick enough that the function of the lobule is preserved, along with the 2-dimensional organization. The number of cortical and medullary regions present in any given slice will vary by the size of the original slice cut. The exact orientation of the tissue being cut does not appear to be critical. This unique property of the thymus likely helps contribute to the ability of the slices to function as a thymus organ.

5. MAJOR MILESTONES

7/6/2018	Module 1 and 4 submitted
12/20/2018	Module 5 submitted
4/5/2019	Module 3 submitted, start of PDFUA clock
6/4/2019	Filing date
9/8/2019	Midcycle meeting
9/27/2019	Late cycle meeting
12/4/2019	PDUFA action date

6. CMC/QUALITY REVIEW TEAM

Reviewer/Affiliation	Section/Subject Matter
Thomas Finn, CBER/ OTAT/DCGT/CTB	1.4.4 – Cross-reference 1.12.5 – Request for a waiver 1.12.14 – Environmental analysis 1.14 – Labeling 1.18 – Proprietary names 3.2.S.1 – General Information 3.2.S.2 – Manufacture 3.2.S.2.4 – Controls of Critical Steps & Intermediates 3.2.S.2.5 – Process Validation 3.2.S.2.6 – Manufacturing Process Development 3.2.S.3 – Characterization 3.2.S.4 – Control of Drug Substance 3.2.P.1 – Description and Composition of the DP 3.2.P.2 – Pharmaceutical Development 3.2.P.3 – Manufacture 3.2.P.3.4 – Controls of Critical Steps & Intermediates 3.2.P.3.5 – Process Validation 3.2.P.5 – Control of Drug Product 3.2.P.6 – Reference Standards or Materials 3.2.A.1 – Facilities and Equipment 3.2.R – Regional Information
Alyssa Kitchel, CBER/ OTAT/DCGT/CTB	3.2.S.6 – Container Closure System 3.2.S.7 – Stability 3.2.P.2.4 – Container Closure System 3.2.P.7 – Container Closure System 3.2.P.8 – Stability
Sukhanya Jayachandra, CBER/ OTAT/DCGT/CTB	3.2.S.2.3 – Control of Materials
Irina Tiper CBER/OTAT/DCGT/CTB	1.14 – Labeling 3.2.S.4 – Control of Drug Substance 3.2.P.4 – Control of Drug Product

7. INTER-CENTER CONSULTS REQUESTED

Reviewer/Affiliation	Section/Topic	In agreement with consult recommendations (Yes/No)
Samanthi Wickramasekara CDRH/OSEL/DBCMS	3.2.P.2 – Final container leachables and extractables	Yes

8. SUBMISSION(S) REVIEWED

Date Received	Submission	Comments/ Status
7/6/2018	125685.0	Original submission with Module 1 and Module 4 information
10/30/2018	125685.1	Proprietary name review
12/20/2018	125685.2	Module 5 rolling submission

4/5/2019	125685.3	Module 3 of rolling submission
4/29/2019	125685.5	Proposed non-proprietary name suffix
5/15/2019	125685.7	(b) (4) and mycoplasma validation
6/14/2019	125685.8	Calculation of tissue slice dose by (b) (4)
6/17/2019	125685.9	Information on 3rd party testing laboratories; (b) (4) facility information; copy of (b) (4) batch record
6/21/2019	125685.10	(b) (4) histology assay method and reference images
6/26/2019	125685.11	Mycoplasma limit of detection validation
6/28/2019	125685.12	Source material hold times; in-process container closures; (b) (4) final container closure, acceptable endotoxin level in materials and final product; drug product batch listing with histology testing times; calculation of residual excipient administered to patient; additional examples of (b) (4) histology from PV lots; donor eligibility; in-house testing of critical materials
7/17/2019	125685.14	Source material (b) (4) storage; culture medium; histology testing time points; justification for culture time window; final product shipping and handling; (b) (4) filters; histology protocol and histological analysis
7/17/2019	125685.15	Facility environmental monitoring; Thymus Organ Media (TOM) medium preparation; (b) (4) validation studies; (b) (4) sampling; facility equipment and procedures; final product visual inspection; aseptic process validation; (b) (4) final container closure; drug product packaging and shipping procedures; container; DP endotoxin levels; DP patient label; DP shipping temperature.
7/18/2019	125685.16	Facility environmental monitoring; (b) (4) sampling;
7/26/2019	125685.17	Manufacturing consistency; batch analysis; clinical experience with (b) (4) container; comparison of (b) (4) final container packaging process; (b) (4) validation; histology assay methods; histology assay validation (b) (4) variability (b) (4)
8/15/2019	125685.18	Process validation; chemokine assays
8/20/2019	125685.19	Donor qualification; donor eligibility
8/23/2019	125685.20	15-day response to 483 observations
8/23/2019	125685.21	Relationship between length of culturing and clinical outcome
8/30/2019	125685.22	(b) (4) sterility validation; endotoxin validation
8/30/2019	125685.24	Donor eligibility
9/6/2019	125685.25	Endotoxin validation; batch records for PV lots (b) (4)
9/13/2019	125685.27	Revised product labeling

9/13/2019	125685.28	Justification for specifications; histology acceptance criteria; histology sample selection; reference histology images; final product visual inspection; Preliminary T cell counts on subjects transplanted with product lots using (b) (4) final container; source material container closures; 6-month T cell counts on subjects (b) (6) who received products lots formulated and stored in (b) (4) final container
9/17/2019	125685.29	Sample (b) (4) for sterility, endotoxin and mycoplasma
9/18/2019	125685.30	Biopsy data on subjects (b) (6) (days 98 and 139), (b) (6)
9/25/2019	125685.32	Dose calculation on PV lots (b) (4)
9/26/2019	125685.33	Cross-reference to IND 9836
10/1/2019	125685.34	Clinical outcome of 7 recently treated subjects
10/1/2019	125685.36	Slicer Performance Qualification Protocol
10/4/2019	125685.37	(b) (4) sterility validation; mycoplasma validation
10/10/2019	125685.39	Donor eligibility and donor qualification
10/15/2019	125685.40	Thymus source material pre-processing and processing hold times
10/15/2019	125685.41	Sterility testing of (b) (4) media
10/15/2019	125685.42	Material qualification sterility and endotoxin testing; (b) (4) sterility assurance method; ancillary material shelf life; facility environmental monitoring; telecon summary of 483 discussions
10/23/2019	125685.43	Results of tissue slicer qualification
10/25/2019	125685.44	Justification for lot size; relationship between surface area of tissue slices and intended dose; proposed labeling information about dose; procedures for ensuring minimum dose and exceeding maximum dose; comparison of dose calculation with (b) (4) methods and historical method
10/28/2019	125685.45	Revised proposal for histology test window; request for extension to update BLA eCTD with (b) (4) final container information
10/28/2019	125685.46	Details of 20 subjects with low or delayed naïve T cell counts
11/1/2019	125685.47	Updated Module 3 with (b) (4) culture dish final container closure
11/1/2019	125685.48	List of treatment related adverse events
11/1/2019	125685.49	2nd response to 483 observations
11/5/2019	125685.50	Additional details on 20 subjects with low or delayed naïve T cell counts
11/7/2019	125685.51	(b) (4) final container closure transport study report
11/12/2019	125685.52	Additional data on subjects with T cell counts (b) (4) and statistical analysis of responders and non-responders

11/15/19	125685.53	Revised package insert in response to first round of FDA feedback
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9. Referenced REGULATORY SUBMISSIONS (e.g., IND BLA, 510K, Master File, etc.)

Submission #	Holder	Referenced Item	Letter of Cross-Reference	Comments/Status
IND 9836	Dr. Mary Louise Markert	The Applicant referenced but did not directly refer to the IND for any specific information. Information from the IND was used by the review team in the preparation of information requests	Submitted 9/26/19 in 125685.3 3	CMC information in IND is at the level of Phase 2 manufacturing. No formal Phase 3 conducted. Patients still being treated under 2 expanded access trials.

10. REVIEWER SUMMARY AND RECOMMENDATION

A. EXECUTIVE SUMMARY

RVT-802 (RETHYMIC) is consists of slices of neonatal allogeneic thymus tissue slices cultured in medium supplemented with (b) (4) FBS for up to 21 days. The product is intended to partially restore thymic function in children born without a thymus due to chromosomal deletions. Very few individuals born with congenital athymia live past age 2. Since the thymus is responsible for T cell maturation of immature T cells homing to the thymus from the bone marrow, and positive and negative selection, patients born without a thymus are severely immunocompromised and have no ability to generate functional naive CD4 and CD8 T cells. Elevated naive T cells can be detected in peripheral blood in transplanted patients beginning around 6 months. The amount of tissue transplanted, even at the maximum dose, represents the 5th percentile of thymus size in children 6 months of age. The level of T cells found years after transplant varies but is typically only about 10% of normal pediatric values. Nevertheless, 72% of transplanted patients live beyond 2 years, with the oldest subject reaching 26 years old.

The therapy was developed under IND 9836 by Dr. Louise Markert at Duke University. The IND was submitted in 2001, but patient treatment with the product and clinical results date back to 1993. Ten different clinical protocols were used under IND and manufacturing information on most of the associated product lots were provided in the submission. Three clinical protocols were used to establish clinical safety and efficacy, and the manufacturing data ranged from 1993 to 2016. The older product lots were manufactured with antibiotics and with deoxyguanosine to diminish the number of allo-thymocytes present in the slices.

The tissue is processed fresh, but can be held (b) (4) at (b) (4). It is sliced (b) (4). Slices are cultured on top of a (b) (4) filter suspended above (b) (4) of culture medium using strips of surgical sponge.

The original submission lacked details and extensive information requests were needed during the review of the BLA. The following major concerns were raised, but addressed by the Applicant:

1. Whether decreased efficacy was seen in subjects transplanted with the product cultured for the full 21 days.
2. Variability of the size and thickness of the cultured tissue slices.
3. Lack of relationship between the amount of starting material and the final lot size.
4. The manufacturing process is not designed to produce the full dose.
5. Whether the use of a more accurate means to measure tissue slice area meant the established product dose was not representative of historical data.

However, unresolved major issues are:

- 1) Samples used for final product release for potency, identity, impurity (residual donor thymocytes), and overall quality are tested up to (b) (4) days prior to drug product formulation, and the product continues to change over time in culture. No repeat testing is performed in the case of a manufacturing deviation.
- 2) The histology assay used for product release is (b) (4) and there is no SOP for histological evaluation performed by the pathologists.
- 3) Portions of the process validation (PV) study are not adequate because of deficiencies in the study design and insufficient information on the properties of the product lots used to support safety and efficacy was provided, thereby making interpretation of the results difficult.
- 4) Insufficient information is available to support the source material holding time of (b) (4) at room temperature prior to processing or further hold (b) (4) at (b) (4).
- 5) Final product expiry of (b) (4) is not supported by available information.
- 6) The original submission proposed to use a (b) (4) specimen container as the final container closure. The change to the (b) (4) container resulted in a change in formulation (b) (4), configuration of the culture slices (b) (4) instead of being suspended above the medium, longer shelf life, differences in product labels and handling procedures by surgical personnel. Proper risk management was not applied, and stability studies were considered insufficient. The Applicant was offered the option of reverting to the (b) (4) final product culture dish container, which Enzyvant chose. A new (b) (4) final container transport study was conducted, but the results were invalid, and thus the (b) (4) container closure has not been adequately validated.

The product is contract manufactured at the (b) (4) manufacturing facility on the (b) (4) campus near the (b) (4). Manufacturing up until 2016 was performed in (b) (4) own manufacturing space. (b) (4) transferred all

manufacturing to the (b) (4) facility and, at that time, a new method for determination of slice surface area (dose) and revised specifications were introduced. The (b) (4) facility is a small multi-product manufacturing space on the (b) (4) of the (b) (4) building, with a dedicated clean room for RETHYMIC product. The inspection of the (b) (4) contract manufacturing facility at (b) (4) resulted eleven 483 observations. The response to the 483 was considered unacceptable. Additional advice was provided to the Applicant in a telecon and the Applicant responded with a second set of responses. Changes made to the quality system were insufficient.

Due to the unresolved product quality and facility concerns, we recommend that the BLA not be approved and that a Complete Response letter be issued.

B. RECOMMENDATION

We recommend that the BLA not be approved and that the following CR items be issued:

I. COMPLETE RESPONSE (CR)

1. You proposed histology-based testing strategy for potency, identity, purity (i.e., safety assessment to evaluate residual donor thymocytes), and overall product quality using product samples collected between Day (b) (4) of manufacturing for the final product (RETHYMIC) released between Day 12-21. Based on these (b) (4) windows, you added a provision that the maximum possible difference between the time of sample collection and time of product release will be (b) (4) days. Per your justification, the primary reasons for performing lot release testing up to (b) (4) days (b) (4) of the drug product formulation are to: 1) accommodate a immunosuppression regimen for some patients using rabbit anti-thymocyte globulin (RATGAM); and 2) have flexibility in the surgical schedule to account for potential delays. The testing strategy is not acceptable because the quality of the final product cannot be adequately evaluated prior to product release and distribution. Please address the following issues:
 - a. We note the following inconsistencies between the proposed testing strategy and your provided justification, particularly with respect to the timeline set for performing release testing.
 - i. You state that there are serious and potentially life-threatening consequences if RATGAM is administered to a patient who is then unable to be treated with RETHYMIC. Both Atypical and typical subjects treated under IND received product lots ranging in culture age between Day (b) (4) Day 21. Since full donor qualification is not known until on or after Day 12 and RATGAM is a three-day course followed by two days of recovery, the earliest a RETHYMIC lot could be administered according to the (b) (4) day test strategy would be Day (b) (4). However, you intend to administer RETHYMIC using lots formulated with cultures harvested as early as Day 12.

- ii. The proposed test window does not accommodate situations in which the patient will not receive RATGAM, a delay in surgery does not occur, or that (b) (4) days is not needed to obtain histology results. For example, a maximum sample collection time point of (b) (4) days is not justified in all cases, and you have not proposed a target for a shorter difference between sampling and release for situations where (b) (4) days are not necessary.
- iii. Endotoxin testing is performed at the time of culture harvest for lot release. If the results are positive, the lot would fail and not be released for administration to the patient. However, no (b) (4) testing is performed before Day 12 in preparation for a possible start of the RATGAM course for the patient.
- iv. No (b) (4) testing is performed to determine the dose, as RETHYMIC dose is calculated (b) (4) before tissue culture harvest and transplantation, regardless of the number or size of the slices generated. The established dose is at least (b) (4) mm² tissue-slice surface area /m² body surface area/, yet the manufacturing process is not designed to assure the minimum dose will always be achieved for each lot manufactured.

The current strategy for performing various tests does not support your justification that lot release testing must be performed (b) (4) by up to (b) (4) days of drug product harvest and formulation. In the interest of maintaining consistently high-quality product for each patient, all lot release testing should be performed on the product at a narrower window of time around or closer to harvest and/or formulation dictated by clinical reasons per a specific patient, rather than reasons arising from scheduling of manufacturing facility, clinical site or staff, and other matters. Please establish a new testing strategy with clear time limits that is fully consistent across all lot release tests you perform on your product.

- b. Please include in your batch record the planned time and the actual time for each lot release test, the harvest, and the transplant. If product harvest and subsequent lot release are delayed for a specific patient because of a clinical reason, such an event needs to be clearly documented in the batch record as being outside of CGMP control.
 - c. You will need to establish procedures for retesting product lots in the event a serious deviation occurs after histology testing has been already conducted. Please establish procedures for performing such retesting with additional slices of tissue and provide relevant Standard Operating Procedures (SOPs).
2. 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 for RETHYMIC evaluation. However, there is no Standard Operating Procedure (SOP) for the procedures performed by the pathologists. 21 CFR 211.100 requires written procedures for both manufacture and process controls designed to assure that the drug products have the identity, strength, quality, and purity. 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 and provides further assurance of product quality. Furthermore, the batch records for histology reported both the (b) (4) results, including reporting a percentage of residual thymocytes. Thus, please revise your histology acceptance criteria to include a (b) (4) measure for assessment of (b) (4).
 - c. Data from twenty-nine patients 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, or at least a two-year delay in development of elevated naïve T cell counts. You have not adequately excluded the possibility that these patients received lower quality lots. You have also not provided examples of histology images from patients who had positive or negative clinical outcomes other than to indicate that histology testing met the release criteria at the time. 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 include examples of the histology images evaluated in your BLA resubmission.
3. You have indicated that incoming thymus tissue can be held for up to (b) (4) at room temperature prior to processing or further hold (b) (4) at (b) (4). Based on the information provided in your process validation (PV) study and previous experience, we have the following concerns:
 - a. Manufacturing instructions for product lots produced under IND in the (b) (4) entailed immediate tissue processing or storage (b) (4) at (b) (4). In addition, you have not provided information that any of the clinical lots used to support safety and efficacy of RETHYMIC were exposed to the full (b) (4) total hold time at room temperature.
 - b. Your 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 under these conditions. The study is insufficient to determine whether the (b) (4) staining 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), stability-indicating 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 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 (b) (4). You have proposed 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) OR hold time.

To support the 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) assay that is stability indicating on multiple lots tested using multiple slices; or establish a shorter expiry based on historical clinical data of safety and efficacy on multiple patients.

4. You have proposed a (b) (4) final product expiration (total time outside incubator until end of surgery) based on one clinical lot manufactured in 2018 that experienced a hold time of (b) (4). However, this data does not support the proposed hold time due to the fact that the RETHYMIC 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 experienced less than (b) (4) to perform all steps through the completion of product administration. Thus, 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.
5. The process validation study does not adequately demonstrate manufacturing and product consistency for all elements. A successful process validation study should demonstrate that each unit operation is performing as intended, and manufacturing is consistent lot-to-lot, but that was not fully demonstrated in Process Validation (b) (4). 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 the presence of key hallmarks of thymus tissue at all stages, the same methods applied to potency and overall tissue quality are not conclusive because:
 - 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 set very broadly, 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 that subjects received and had either a positive outcome, negative outcome, or a reduced/delayed naïve T cell response).
 - 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 a consequence of 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 effort to cover the range of conditions the lots would be exposed to for commercial manufacturing, typically, PV is performed after critical process parameters for all manufacturing steps have been established. It is customary that for process validation a minimum of three lots be manufactured under the same conditions. The information provided in this study is useful, but more 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 a single lot. Further, the study was designed to use only a single lot of culture medium per lot, yet most clinical lots used multiple, and you report for some clinical lots used as many as (b) (4). 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 for verification.
 - iii. The clinical data set indicates that about (b) (4) of slices produced with the tissue slicer are thick and (b) (4) are thin, though the proportion varies by 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.
- 6. Transport study, (b) (4)-2019-050-A, provided in Amendment 51 (received on 11/7/19), was conducted to validate that the (b) (4) final product container maintains a sterile

environment during transport from the (b) (4) facility to the operating room (OR). Based on the results provided, this transport study is currently inconclusive, because it has failed to demonstrate positive growth in medium growth promotion tests. As stated in Amendment 51, you intend to investigate the source of these failure and submit a final report upon completion of the investigation. Please provide this final report that include details of this investigation and source of these failures, root cause, and appropriate corrective actions. Additionally, please provide a transportation study that demonstrates the final drug product container adequately maintains a sterile environment during transport from the (b) (4) facility to the OR.

II. SIGNATURE BLOCK

Reviewer/Title/Affiliation	Concurrence	Signature and Date
Thomas Finn, CBER/OTAT/ DCGT/CTB	Concur	Thomas P. Finn -S 2019.11.26 16:10:16 -05'00'
Alyssa Kitchel, CBER/OTAT/ DCGT/CTB	Concur	Alyssa Kitchel -S 2019.11.26 16:46:28 -05'00'
Sukhanya Jayachandra, CBER/OTAT/DCGT/CTB	Concur	Sukhanya Jayachandra -S <small>Digitally signed by Sukhanya Jayachandra -S DN: c=US, o=U.S. Government, ou=FDA, ou=People, 0.9.2342.19200300.100.1.1=1300391929, cn=Sukhanya Jayachandra -S Date: 2019.11.26 16:46:28 -05'00'</small>
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TABLE OF CONTENTS

Module 3	4
3.2.S DRUG SUBSTANCE	4
3.2.S.1.1 - 1.3 Nomenclature, Structure and General Properties	4
3.2.S.1.1 Nomenclature	4
3.2.S.1.2 Structure	4
3.2.S.2 Manufacture	5
3.2.S.2.1 Manufacturer(s)	5
3.2.S.2.2 Description of Manufacturing Process	5
3.2.S.2.3 Control of Materials	11
3.2.S.2.4 Controls of Critical Steps and Intermediates	14
3.2.S.2.5 Process Validation and/or Evaluation	32
3.2.S.2.6 Manufacturing Process Development	44
3.2.S.3 Characterization	45
3.2.S.3.1 Elucidation of Structure and Other Characteristics	45
3.2.S.3.2 Impurities	45
3.2.S.4 Control of Drug Substance	45
3.2.S.4.1 Specification(s) and 3.2.S.4.5 Justification of Specification(s)	45
3.2.S.4.2 Analytical Procedures and 3.2.S.4.3 Validation of Analytical Procedures	52
3.2.S.4.4 Batch Analyses	58
3.2.S.5 Reference Standards or Materials	59
3.2.S.6 Container Closure System	59
3.2.S.7 Stability	61
3.2.S.7.1 Stability Summary and Conclusion and 3.2.S.7.3 Stability Data	61
3.2.S.7.2 Post-Approval Stability Protocol and Stability Commitment	63
3.2.P DRUG PRODUCT	64
3.2.P.1 Description and Composition of the Drug Product	64
3.2.P.2 Pharmaceutical Development	64
3.2.P.2.1 Components of the Drug Product	64
3.2.P.2.3 Manufacturing Process Development	64
3.2.P.2.4 Container Closure System	64
3.2.P.2.5 Microbiological Attributes	64
3.2.P.3 Manufacture	64
3.2.P.3.1 Manufacturer(s)	64
3.2.P.3.2 Batch Formula	65
3.2.P.3.3 Description of Manufacturing Process	65
3.2.P.3.4 Controls of Critical Steps and Intermediates	66
3.2.P.3.5 Process Validation and/or Evaluation	66
3.2.P.4 Control of Excipients	66
3.2.P.4.5 Excipients of Human or Animal Origin	67
3.2.P.4.6 Novel Excipient	67
3.2.P.5 Control of Drug Product	67
3.2.P.5.1 and 3.2.P.5.6 Specification(s) and Justification of Specification(s)	67
3.2.P.5.2 and 3.2.P.5.3 Analytical Procedures and Validation of Analytical Procedures	68

3.2.P.5.4 Batch Analyses.....	69
3.2.P.5.5 Characterization of Impurities.....	69
3.2.P.6 Reference Standards or Materials	69
3.2.P.7 Container Closure System.....	69
3.2.P.8 Stability.....	74
3.2.P.8.1 Stability Summary and Conclusion and 3.2.P.8.3 Stability Data	74
3.2.P.8.2 Post-Approval Stability Protocol and Stability Commitment	75
3.2.A APPENDICES.....	75
3.2.A.1 Facilities and Equipment	75
3.2.A.2 Adventitious Agents Safety Evaluation	76
3.2.A.3 Novel Excipients	78
3.2.R Regional Information ((b) (4) facility).....	78
Module 1	80
A. Environmental Assessment or Claim of Categorical Exclusion	80
B. Labeling Review.....	80
Modules 4 and 5.....	81
Analytical Procedures and Validation of Analytical Procedures for Assessment of Clinical and Animal Study Endpoints	81

TABLE OF FIGURES

Figure 1: Manufacturing overview.	i
Figure 2: Organization of the thymus and role of thymus in T cell maturation.	ii
Figure 3: Manufacturing flowchart.	6
Figure 4: Overview of manufacturing and testing.	7
Figure 5: Relationship between sampling for histology and release of product.	8
Figure 6: Total dose produced per lot as a function of (b) (4) of source material.....	21
Figure 7: Depiction of filter coverage based on our analysis of provided data at the beginning and end of culture period.	22
Figure 8: Slice size distribution among lots	23
Figure 9: Relationship between the number of slices produced and the total dose transplanted.	24
Figure 10: Scatterplot of BSA Versus Dose for Lot Sizes of (b) (4) Slices	25
Figure 11: Depletion of thymocytes between days (b) (4) of culture.....	25
Figure 12: Composite images demonstrating the presence of non-viable and potentially viable T cells in Day (b) (4) cultures (left panels). Presence of a large amount of (b) (4) staining (right panel).....	26
Figure 13: Histological appearance of Day (b) (4) and Day 21 tissue.	26
Figure 14: Typical (b) (4) staining on Day (b) (4)	27
Figure 15: Changes in (b) (4) staining throughout culture.....	27
Figure 16: Days in culture and number of slices transplanted.	28
Figure 17: Culture age at the time of transplant.....	29

Figure 18: Tissue (b) (4) in culture.....	30
Figure 19: Representative images from time course series.....	37
Figure 20: Comparison of time course of PV lots (b) (4) by (b) (4) staining.....	38
Figure 21: Representative fields of view of the same histology section depicting widely different morphology of (b) (4) expression.....	39
Figure 21: Time course of (b) (4) expression from selected additional images provided in response to an information request.	40
Figure 23: Enlargement of Day 21 staining of (b) (4)	41
Figure 24. Examples of biopsies from two subjects treated under IND 9836 demonstrating.	43
Figure 24: (b) (4) facility floorplan.....	76
Figure 25: (b) (4) culture dish label.....	80
Figure 26: (b) (4) acrylic container label.	81

TABLES

Table 1: RETHYMIC drug substance manufacturing and testing sites.....	5
Table 2: Product manufacturing differences throughout product history.....	9
Table 3: Reviewer evaluation of control strategy.....	15
Table 4: Batches using less than (b) (4) of starting material.	20
Table 5: PV times for manufacturing steps.....	33
Table 6: Dose by responder status at Years 1 and 2.	42
Table 7: Evaluation of drug substance and drug product release specifications.	46
Table 8. DS holding times during manufacturing process.	62
Table 9: Average, maximum, and minimum hold times for the (b) (4) hold step....	62
Table 10: DP manufacturing and testing sites.....	64
Table 11: Bath formula of RETHYMIC.	65
Table 12: RETHYMIC drug product excipients.....	66
Table 13: Specifications for RETHYMIC drug product.	67
Table 14. Summary of DP final container changes from the IND to the original BLA submission to the amended BLA submission (Amendment 49).....	70
Table 15: Assays used for donor eligibility testing.....	77
Table 16: Additional thymus donor testing.	78

Module 3

3.2.S DRUG SUBSTANCE

3.2.S.1.1 - 1.3 Nomenclature, Structure and General Properties



3.2.S.1.1 Nomenclature

Company assigned name: RVT-802

Non-proprietary name: RETHYMIC

Drug substance lots are assigned a manufacturing lot number (b) (4)-XXX, where X is a number, and given an ISBT product lot number.

(b) (4)



3.2.S.2 Manufacture

3.2.S.2.1 Manufacturer(s)

(b) (4)

3.2.S.2.2 Description of Manufacturing Process

(b) (4)

58 pages determined to be not releasable: (b)(4)

3.2.P DRUG PRODUCT

3.2.P.1 Description and Composition of the Drug Product

The drug substance and drug product are essentially the same. Each drug product container contains up to four slices of thymus tissue, each adhered to a filter placed on top of a surgical sponge in 5 mL of thymus organ media (TOM). Each container includes up to 2 surgical sponges and 2 tissue slices adhered to filters that are placed on top of each surgical sponge with up to 4 tissue slices per container. The same media ingredients that are used to culture the drug substance are included with the drug product. The container is the same 100 mm diameter polystyrene culture dish with lid that is used throughout culture of the drug substance. Up to 11 containers are supplied to the operating room for treating each patient.

3.2.P.2 Pharmaceutical Development

3.2.P.2.1 Components of the Drug Product

The drug substance contains the same ingredients as the drug product, with the use of the same media components, filter, surgical sponge and container closure.

3.2.P.2.3 Manufacturing Process Development

The manufacturing process is uninterrupted from initiation of tissue slice to formulation and packaging of the DS into the DP. Please see Section [3.2.S.2.2](#) and [3.2.S.2.6](#) for the developmental history.

3.2.P.2.4 Container Closure System

The RETHYMIC thymus tissue slices are supplied in the same cell culture dish with lid that is used for the drug substance. Please see [Section 3.2.P.7](#).

3.2.P.2.5 Microbiological Attributes

Microbiological testing of RETHYMIC thymus tissue slices is performed as described in [Section 3.2.S.4.1](#).

3.2.P.3 Manufacture

3.2.P.3.1 Manufacturer(s)

Table 10: DP manufacturing and testing sites

RETHYMIC Drug Product Manufacturing and Testing Sites	
Manufacturing Site	Responsibilities
(b) (4)	

Testing sites for DP release	Responsibilities
(b) (4)	

3.2.P.3.2 Batch Formula

Table 11: Bath formula of RETHYMIC.

Component	Maximum quantity per batch
RETHYMIC	(b) (4) slices
(b) (4) filter	(b) (4) filters
Surgical sponge	(b) (4) sponges
TOM	
(b) (4)	(b) (4)
(b) (4)	(b) (4)
(b) (4)	(b) (4)
FBS	(b) (4)
Total	(b) (4) of TOM with (b) (4)

3.2.P.3.3 Description of Manufacturing Process

The manufacturing process is summarized in Figures 1,3, and 4 and referenced throughout the memo. Drug product testing includes testing performed on (b) (4). The DP is generated on the day of transplant when the product is formulated and labeled. The excipients are the same as the culture medium used for (b) (4) manufacture except only 5 mL is used for formulation (instead of (b) (4) used in culture). The DP container closure is the same (b) (4) container closure used (b) (4). The cultures are technically not harvested because no manipulation is performed on the slices and the slices are not combined in any way, other than how organized during (b) (4) manufacture. Not indicated in Figures 1-4 is that after formulation the DP is placed back in the incubator until the

operating room calls to inform the (b) (4) facility the patient is under anesthesia and the surgeon is ready to receive the product. At that point the DP culture dishes are removed from the incubator, the final product label is applied to the primary and secondary containers and the expiration is calculated. Transport to the OR typically takes (b) (4). Once in OR the product is either transplanted right away, or after completion of another surgical procedure. Although the placement of the DP back into the incubator represents both a further culture step and a second storage condition, the length of time is short. The Applicant should establish a step time for this phase of manufacturing which should be part of the control of the DP and included in future process validation studies (see [3.2.S.2.5](#)). It is not expected that the incubator step would change the product in any significant way, and should help maintain product stability.

3.2.P.3.4 Controls of Critical Steps and Intermediates

Because the generation of the DP is so closely associated with the DS, additional in-process testing of the DS is not necessary. The final media exchange representing formulation is nearly identical to all the (b) (4) media exchanges performed on the DS. The hold time for the short time that the DP is held in the incubator prior to transport to the OR is not captured. A step time should be established and included as part of the control of process and process validation. Packaging procedures are covered by SOP. (b) (4) staff evaluate by visual inspection the transported DP inside the OR. The critical quality attributes (CQAs) for the full manufacturing process are provided in [Section 3.2.S.2.4](#).

3.2.P.3.5 Process Validation and/or Evaluation

Most process validation studies are described in [Section 3.2.S.2.5](#), since the drug product is essentially the same as the drug substance.

3.2.P.4 Control of Excipients

Excipients for the drug product are the same components of the DS culture medium. The only difference is that the (b) (4) used during the 21-day culture. Excipients are added at the time of final product preparation. Afterwards, the culture plates are placed back in the tissue culture incubation until the surgical team calls (b) (4) in advance of when they want the product delivered to the OR.

Table 12: RETHYMIC drug product excipients.

Component	Function	Amount/Concentration
Thymus Organ Media (TOM):		5.0 mL
(b) (4)		
(b) (4)		
(b) (4)		
Fetal Bovine Serum (FBS)	Growth supplement	(b) (4)

Reviewer comments: In the original submission the Applicant listed the surgical sponge and (b) (4) filter as components of the final product, but did not list them as either part of the container closure or as an excipient. They would have to be one or the other. This was brought to the Applicant's attention and the Applicant responded in Amendment 14 that because the filters and sponge are used as processing aids, they do not readily fall into either category of excipient or container closure system. The review team acknowledges that these materials are atypical drug components, but disagree with their conclusion. In discussion with DCGT management and DMPQ review staff, we determined that the appropriate category should be part of the container closure system and not excipients for the following reasons: 1) neither the filter or the sponge are transplanted in the patient; and 2) both are present to provide physical support for the tissue slices, which fits within the definition in guidance of a container closure system. The revised eCTD submission provided in Amendment 47 now lists the surgical sponge and filter as part of the container closure system.

The (b) (4) sponge is supplied by (b) (4) and meets the requirements for (b) (4). It is a (b) (4) that is intended for hemostatic use.

The (b) (4) sterile filters are (b) (4) made of (b) (4). Porosity is rated as (b) (4). They are compatible with (b) (4) sterilization methods. They are intended for analytical and research applications.

For additional details on the validation of the sponge and filter, please see [Section 3.2.S.6](#).

3.2.P.4.5 Excipients of Human or Animal Origin

The list of excipients includes (b) (4) FBS. (b) (4) FBS is used for DS manufacturing and DP formulation. Please see [Section 3.2.S.2.3](#).

3.2.P.4.6 Novel Excipient

The surgical sponge and (b) (4) filter are categorized as part of the container closure.

3.2.P.5 Control of Drug Product

3.2.P.5.1 and 3.2.P.5.6 Specification(s) and Justification of Specification(s)

Table 12 outlines the tests performed on the DP. The DP is released without available sterility and mycoplasma results. Visual inspection, endotoxin, and (b) (4) results are available for release. For sterility, all (b) (4) are (b) (4) or reported immediately if the product becomes positive. Thus, the DP is release based on a negative (b) (4) sterility result and (b) (4).

Table 13: Specifications for RETHYMIC drug product.

Attribute	Timing of test and result availability	Test Parameter and Test Method	Acceptance Criteria
Identity	Final product container is examined and results are immediately available.	Visual inspection of the container	<ul style="list-style-type: none"> No evidence of tampering or damage to containers Yellow to brown slices of tissue with varying thickness and shape, adhered to round white membrane filter (b) (4)
Safety	DP testing is performed on the day of release to obtain results prior to release.	(b) (4)	(b) (4)
Safety	The lot is released if DS results are negative, but prior to receipt of result obtained on the day of transplant.	(b) (4)	No growth
Safety	DP results are not available for release. (b) (4)	Sterility via (b) (4)	No growth
Safety	(b) (4) is collected on the day of transplant. DP results are available for release.	(b) (4)	(b) (4)

Overall Reviewer's Assessment of Sections 3.2.P.5.1 and 3.2.P.5.6:

These sections are adequate. The risk assessment related to the timing of sterility, endotoxin, and mycoplasma testing is deferred to OCBQ/DBSQC. DBSQC recommended approval of the methods. Please refer to the review memo by Simleen Kaur.

3.2.P.5.2 and 3.2.P.5.3 Analytical Procedures and Validation of Analytical Procedures

Identity testing is the only DP release test. Visual inspection of the drug product for appearance of container integrity and tissue slices is performed on the day of release to confirm the following:

- No evidence of tampering or damage to containers.
- Yellow to brown slices of tissue with varying thickness and shape, adhered to round white membrane filter.

- (b) (4)

Identity testing conducted on the DP immediately before release does not require method validation.

In the original BLA, the Applicant did not include a criterion for visual inspection of the media. The Applicant was advised that visual inspection of the final product in the final container should include an evaluation of the appropriate (b) (4) of medium and evaluate for the presence of (b) (4) (IR sent August 31, 2019). The Applicant adequately revised the acceptance criteria for visual inspection in Amendment 30 (dated September 13, 2019).

Overall Reviewer's Assessment of Sections 3.2.P.5.2 and 3.2.P.5.3:

These sections are acceptable.

3.2.P.5.4 Batch Analyses

The Applicant submitted data on batch size surface area and number of slices (batch size) for lots (b) (4). The acceptance criteria for PV lot surface area was (b) (4) mm²/m² BSA. The lot surface area for the commercial product is (b) (4)-22,000 mm²/m² BSA. The Applicant's manufacturing experience suggests that the dosage specification can be met.

3.2.P.5.5 Characterization of Impurities

The impurities in the RETHYMIC drug product are the same as in the RETHYMIC drug substance, as described in Section [3.2.S.3.2](#).

Overall Reviewer's Assessment of Sections 3.2.P.5.4 and 3.2.P.5.5:

These sections are adequate.

3.2.P.6 Reference Standards or Materials

Reference (b) (4) of histology sections that represent acceptable histology for release have been established. See [Section 3.2.S.4.2](#).

3.2.P.7 Container Closure System

The primary container closure initially proposed in the BLA submission was a 100 mL polypropylene specimen container with a polyethylene screw cap from (b) (4). In Amendment 49 (submitted on 10/31/2019), the Applicant provided updated eCTD information to change the container from this (b) (4) container and revert back to the polystyrene (b) (4) tissue culture dish used in the manufacturing of the product. Table 14 summarizes of the DP container changes from the IND to the BLA.

Reviewer comments: The use of this container limits the Applicant's manufacturing and implementation to a single site (b) (4) since it is not suitable for shipping farther than currently tested.

(b) (4)

Reviewer comments: Acceptance criteria for the endotoxin testing was not specified. This issue should be followed up on during review of the CR response. As for the (b) (4) level of the final DP container, cell therapy products that are cultured on tissue culture polystyrene are most likely to be cultured on plates that have an (b) (4) (b) (4) Based on the historical use of these plates in this IND and overall in the field, this (b) (4) appears to be adequate. However, CMC defers to DMPQ on their assessment of the (b) (4) for this final DP container.

(b) (4) to DP upon receiving a call from the OR about the surgery (approximately (b) (4) before). The tissue dishes are removed from the incubators, (b) (4) TOM is added to the tissue culture dish (5 mL). Once the media is changed, the dishes are put back in the incubator until notification is received from the OR.

Reviewer Comment: In the IND, the OR notification was received once the patient was under anesthesia. In the most recent transport study (Amendment 51) the dishes were put back in the incubator for (b) (4) after media change to mimic the packaging and transport.

Each tissue culture dish contains 4 tissue slices, 2 per sponge. A maximum of eleven DP containers can be transported to the OR. These containers are stacked into (b) (4) container - single-use polycarbonate container system (b) (4) which has a capacity of (b) (4) tissue culture dishes. The container contains a silicone gasket seal on the container lid, which is attached to the container tray via 4 polycarbonate latches. This secondary container is then placed into an insulated shipping container (b) (4) which is further insulated with insulation packs. Prior to its initial use, the shipping container is given an identification number and qualified via a thermal quality test. Prior to each use, the insulated shipping container is wiped clean (b) (4)-THY-009 FRM1).

In the original submission, product shipping validation report (b) (4) was provided to demonstrate that the final product could be transported from the (b) (4)

Reviewer comments: The adequacy of the thermal quality test from the original PV study was determined by DMPQ. Please refer to DMPQ's memo.

The E&L testing performed on the (b) (4) closure will be leveraged for the DP container closure, (b) (4). Thus, please refer to the E&L discussion in [Section 3.2.S.6](#) for details regarding such testing.

Due to the changes in the primary container closure, a revised shipping study was conducted, for which the interim report (b) (4)-2019-050-A) was submitted in Amendment 51 on November 7, 2019. This revised study was designed to show that the (b) (4) tissue culture dish could maintain a sterile environment for the DP as it was transported from the (b) (4) facility to the OR. This study utilized (b) (4) media (microbiological media) in each of the 11 DP containers as (b) (4) facility technicians (b) (4) the shipping container to the OR at Duke Hospital. (b) (4) different shelf life scenarios were simulated for three different shipments – (b) (4). During the shipment simulations, the shipping container was taken to the OR, returned to the (b) (4) facility, and then held in the at the facility at room temperature until the total shelf life scenario elapsed. At the end of the shipment simulations, (b) (4) media was collected in (b) (4) and then shipped to (b) (4) for testing. Acceptance criteria was set at no microbial growth per (b) (4) on all samples.

Overall, study results were submitted as an interim report. The test results of the (b) (4) samples from the transport study showed no microbial growth across (b) (4) organisms: (b) (4)

However, for growth promotion stage, the test resulted in two failed samples – no growth, thus the results are considered invalid. Additional (b) (4) samples were retested for growth promotion, which again resulted in an invalid test (b) (4) samples showed no growth). An investigation of the source of these failures is ongoing and the Applicant states that they will provide a final report following completion of the investigation.

Overall Reviewer's Assessment of Section 3.2.P.7:

The information provided to support the proposed (b) (4) tissue culture dish container is not adequate.

The late change in container back to the original primary container used in the clinical studies from 1993 – 2018 is generally supported by the decades of historical clinical data. In addition to the historical experience, it was necessary for the Applicant to conduct an additional transport study to demonstrate that the primary container closure is able to prevent microbial contamination of the DP during transport to the OR. No such study had been performed previously.

Unfortunately, the invalid test results due to the lack of microbial growth in growth promotion conditions cannot adequately support the use of this primary container closure at the time. The investigation as to the source of the growth promotion failures are currently being conducted. The Applicant is considering the failure source to be due to a shorter growth period than specified (b) (4)

Thus, the primary container closure will be a CR item due to the invalid test results. Please see [CR item # 6](#).

3.2.P.8 Stability

3.2.P.8.1 Stability Summary and Conclusion and 3.2.P.8.3 Stability Data

As stated previously, no ICH stability studies were conducted for the drug product. A stability study was conducted as part of the PV using the (b) (4) Specimen Container described previously in [Section 3.2.P.7](#). However, the PV runs to assess DP hold times submitted in the original BLA are no longer applicable due to the change in DP container closure (Amendment 49).

In Amendment 49 and 53, the Applicant proposed that the DP shelf life or “hold time” be limited to (b) (4). This hold time is the total time outside the incubator to the end of surgery and is based on the clinical experience for lots manufactured in both the (b) (4) and the (b) (4) facility. The maximum hold time for the DP recoded by the (b) (4) using the (b) (4) tissue culture dish was (b) (4) (OR arrival time to end of surgery), while the (b) (4) facility recorded a maximum of (b) (4) hold time.

Reviewer Comment: In the revised Module 3 sections included in Amendment 47 establishing the (b) (4) culture dish as the final container the Applicant provided additional data on surgical times. Throughout the review process, including pre-BLA discussions, there was an understanding that the use of the (b) (4) container would involve a longer surgical time because of the additional number of containers and handling procedures. At the Late Cycle Meeting the Applicant clarified that the surgeries take the same length of time regardless of container based on direct observation of each in the OR. The surgeries take about an hour and half. However, the updated information included in [Section 3.2.P.2.3](#) stated that transplant of RETHYMIC was not always the only surgery being performed on the subject. For some subjects another surgery was performed first, thereby extending the time the final product is in the OR. The actual RETHYMIC transplant surgery time was not always recorded. Typically, the delay was only about (b) (4) (time of thymus surgery start relative to product delivery to OR) but was as long as an additional (b) (4). The maximum time of (b) (4) in the OR was due to another surgical procedure being performed first.

Based on information from the IND regarding clinical lots manufactured in the Markert facility, time prior to arrival in the OR could be estimated to be (b) (4). This includes (b) (4) from notification from OR to start of transport and (b) (4) to (b) (4) the DP to the OR. Thus, estimated worst case for the (b) (4) could be over (b) (4)

The Applicant bases their justification for (b) (4) on one clinical lot manufactured in 2018 that had a total hold time of (b) (4). The subject treated with this lot has had a 6-month follow up and is demonstrating thymic function.

Reviewer comments: This one lot with the (b) (4) hold time does not support the proposed hold time due to the fact that it was shipped in the (b) (4) container, not the (b) (4) tissue culture dish.

A (b) (4) study was conducted and described in Section 3.2.S.3 (Section 1.4). This study (b) (4) at day (b) (4) not between days 12-21 when DP would be harvested, to demonstrate the tissue's ability to (b) (4)

. Overall this study does not reflect stability of the DP as it relates to shelf life, but rather (b) (4) stability during manufacturing.

3.2.P.8.2 Post-Approval Stability Protocol and Stability Commitment

As previously stated in Section [3.2.S.7.2](#), no post-approval stability commitment was included in the BLA package due to the fact that the stability of the DP on the order of hours. The Applicant should provide a post-approval stability protocol and commitment to be reviewed as part of the BLA.

Overall Reviewer's Assessment of Section 3.2.P.8:

The information provided to support the proposed (b) (4) shelf life of the final product is not adequate.

The clinical lots that can adequately support a proposed hold time are ones that were transported using the (b) (4) tissue culture dish, and for which there is long term clinical data (> 2 years) that demonstrates the tissue functionality in vivo. This in vivo functionality is important since the only assessment performed on the tissue is histology, which has been a review issue (see Section [3.2.S.4](#)). Furthermore, in the absence of a (b) (4) stability-indicating assay, we can only evaluate the tissue quality and function from historical clinical outcomes.

If the applicant qualifies and validates a (b) (4) assay to assess final product stability over the proposed (b) (4), such an assay could help support this extended shelf life.


Thus, a CR comment will be issued to the Applicant regarding product stability. Please see [CR item #4](#).

3.2.A APPENDICES

3.2.A.1 Facilities and Equipment

(b) (4)

(b) (4)

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(b) (4)

3.2.A.2 Adventitious Agents Safety Evaluation

This section describes the review of procedures for donor eligibility determination and tracking of RETHYMIC manufactured using thymus tissue obtained from allogenic unrelated donors under the age of 9 months who are undergoing cardiac surgery.

Birth mothers and infant donors are evaluated for relevant communicable disease agents or diseases (RCDADs) in accordance with 21 CFR part 1271, subpart C. Tables 14 and 15 outline the testing and screening performed. Thymus tissue from eligible donors and meeting other pre-established criteria qualify for licensure.

Based on the information received and reviewed by Safa Karandish (CBER/OTAT/DHT/HTRB), the Applicant's procedures for donor eligibility determination and tracking are acceptable.

Table 15: Assays used for donor eligibility testing.

Assay	Test kit	Lab
Cytomegalovirus (CMV) Antibody	(b) (4)	(b) (4)
Hepatitis B Surface Antigen (HBsAg)		
Hepatitis B core (HBc) Antigen Antibody		
Hepatitis C Virus (HCV) Antibody		
Human Immunodeficiency Virus (HIV)-I/II/O Antibodies		
Human T-Lymphotropic Viruse (HTLV)-I/II Antibodies		
Treponema pallidum Antibody		
Nontreponemal Syphilis Screen		
Trypanosoma cruzi (chagas)		
HIV-I/II/O / HCV / HBV (b) (4)		
HIV-I/II/O discriminatory (b) (4)		
HCV discriminatory (b) (4)		
HBV discriminatory (b) (4)		
West Nile Virus (b) (4)		
Zika Virus (b) (4)		

Additional testing is performed beyond what is required by regulation and FDA guidance because the patients are immunodeficient, and the risk any infectious agent might pose to the recipient. Not all additional testing involves infectious agents, but additional infectious agent testing involves either repeat testing for additional confidence of the results, or to evaluate viruses that are not required by regulations but could pose a risk to patients. Other tests, such as HLA typing are conducted because HLA matching, though not typically performed, is described in the package insert. (b) (4) analysis to detect a chromosomal analysis is conducted to confirm that the donor does not have DiGeorge, and flow cytometry to confirm that T cells counts and ratios are normal (demonstrating normal thymic function).

Of all these tests, the repeat CMV test is most important. CMV infection in this population is fatal. The donor eligibility procedures the Applicant uses allows for the

mother of the infant donor to be positive for CMV if the recipient tests negative. This is an acceptable procedure if the assay is validated. The CMV assay was validated and was reviewed as part of the (b) (4)

The (b) (4) is in the same building as the (b) (4) facility.

Table 16: Additional thymus donor testing.

Assay	Source	Lab
(b) (4) for 22q11 deletion	Blood Thymocytes	(b) (4)
Lymphocyte Enumeration (Flow Cytometry)	Blood	
CMV PCR (b) (4)	Blood	
Toxoplasmosis (b) (4)	Blood	
(b) (4)		
EBV (b) (4)	Blood	
Blood type	Blood	
Human leukocyte antigen (HLA)	Blood, Thymocytes	
Complete blood count (CBC) with differential	Blood	

□ Viral Clearance Studies

Not applicable

Overall Reviewer's Assessment of Section 3.2.A.2:

As reviewed by Safa Karandish (CBER/OTAT/DHT/HTRB), the Applicant's procedures for donor eligibility determination and tracking are acceptable. Additional test procedures were also reviewed and found acceptable. Donor eligibility determination was also review on inspection an no deficiencies were found.

3.2.A.3 Novel Excipients

Not applicable

3.2.R Regional Information (b) (4) facility)

□ Executed Batch Records

An executed batch record for process validation lot (b) (4) was included in the original submission. Additional batch records were provided through information requests. These include all three PV lots, (b) (4), and three recent clinical lots (b) (4). All three clinical lots were formulated and shipped using the (b) (4) final container.

Batch records (b) (4) include some modified procedures to accommodate the required PV time course studies, such as (b) (4). (b) (4) were the first three lots product lots where the (b) (4) container was used as the final container. Though no batch record was provided for a clinical product lot that matches the proposed commercial process, most of the procedures and associated SOPs are the same as for the (b) (4) lots.

All (b) (4) batch records were reviewed on inspection. Deviations associated with (b) (4) were noted and are included in issued 483 observations. For review see EIR, Amendments 20 and 42, and DMPQ review.

□ **Method Validation Package**

A tissue slicer qualification protocol was provided in Amendment 36, and the results of the study provided in Amendment 43. For review of the tissue slicer study please refer to the DMPQ facility review.

□ **Combination Products**

Not applicable

□ **Comparability Protocols**

The Applicant did not include a comparability protocol, and no ongoing activities at this time require comparability. However, future activities may require a comparability study be performed, including the following possible changes the Applicant is considering:

- As discussed in [Section 3.2.P.7](#), neither the initially-proposed (b) (4) final product container, nor the (b) (4) culture dish (b) (4) DP container are adequately supported. The Applicant will need to identify an appropriate container and provide additional validation data. Depending on the nature of the change a comparability study may be needed.
- (b) (4)

(b) (4) Any additional location for source material supply or product manufacturing will require a comparability study. No other manufacturing sites have been established. To meet patient demand, additional

sites will likely be needed. We strongly recommend that the Applicant develop a comparability plan to meet future needs.

- Depending upon the approach taken by the Applicant to address CR items, a (b) (4) stability indicating assay may be required. A comparison of the new assay with existing assay will be needed, and the introduction of such an assay could be part of a comparability protocol. Given the complexity of the product, the limited control strategies in place, and the deficiencies of the PV study ([Section 3.2.S.2.5](#)), we recommend that the Applicant provide a comparability protocol as an amendment to the BLA prior to initiating any substantial manufacturing change.

Module

A. Environmental Assessment or Claim of Categorical Exclusion

The Applicant is claiming a categorical exclusion under 21 CFR 25.31 (c) from the need to prepare an environmental assessment.

B. Labeling Review

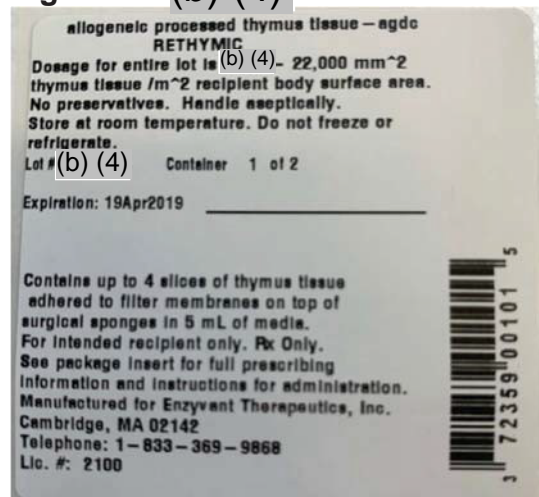
Full Prescribing Information (PI):

The Applicant did not submit a revised Prescribing Information label after the conversion from the (b) (4) container to (b) (4) culture dish (please see Amendment 47 dated 11/01/19). Therefore, this section could not be reviewed.

Carton and Container Label:

A full review of the final container label will occur in the BLA resubmission. It is unclear what final container will be used and what the formulation and expiry will be. No major issues were found in the initial review of the final container and secondary container labels. It is recommended that the final container for any formulation of RETHYMIC include instructions not to agitate, as that could damage the slices adhered to the filters.

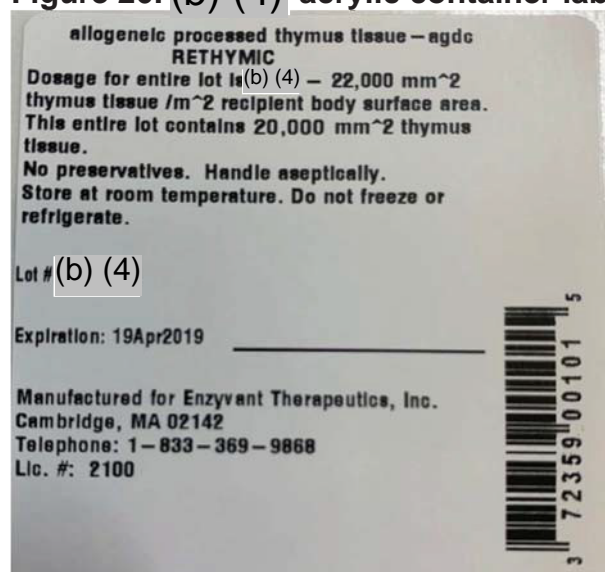
Figure 25: (b) (4) culture dish label.



Up to (b) (4) culture dishes are used during production of the (b) (4). Up to (b) (4) dishes will be used to for the Drug Product. Four culture slices are present in each dish. The surface area of the slices in each dish is calculated on a per dish, not per slice basis. Dishes are numbered in the order they were originally prepared. No numbers are assigned to individual slices. The expiration date is assigned at the time of Drug Product formulation. All tissue slices are assigned the same expiry. The final product is placed back in the tissue culture incubator prior to shipment. Instructions to the surgical team as to how many dishes of DP constitute the minimum dose is communicated by (b) (4) facility present in the OR and responsible for handling the product.

The secondary (b) (4) acrylic container is labeled as follows:

Figure 26: (b) (4) acrylic container label.



A single (b) (4) secondary container can hold up to (b) (4) culture dishes, so only one secondary container is needed. No major issues were found with a preliminary review of the secondary container label.

Modules 4 and 5

Analytical Procedures and Validation of Analytical Procedures for Assessment of Clinical and Animal Study Endpoints

Preclinical data is not relevant for this file. The clinical studies began eight years before IND submission in 2001.

Clinical data from Module 5 containing information about the relationship between product properties and the lack of clinical outcome was reviewed and is discussed in [Sections 3.2.S](#) and [3.2.P](#).

Manufacturing information such as slice size, thickness, total surface area, recipient body surface area, dose, immunosuppression regimen, and other parameters was captured in a spreadsheet and used for CMC analysis. The major findings are reported in [Sections 3.2.S.2.4](#) and [3.2.S.2.5](#). Biopsy information was requested as an Amendment and is discussed in [Section 3.2.S.2.5](#)