

CBER CMC BLA Resubmission Review Memorandum

BLA STN 125706

Remestemcel-L-rknd

Reviewer/Title/Affiliation

Heba Degheidy, MD, PhD | Staff Fellow | CBER/OTP/OCTHT/DCT1/CTTB
Carolina Panico, MD, PhD | Biologist | CBER/OTP/OCTHT/DCT2/TEB2
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1. **BLA#:** STN 125706; Complete Response submitted in 125706/0.65, SN0065

2. **APPLICANT NAME AND LICENSE NUMBER**

Mesoblast, Inc.

3. **PRODUCT NAME/PRODUCT TYPE**

USAN/Proper/Non-Proprietary Name: Remestemcel-L-rknd

Proprietary Name: RYONCIL

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

Remestemcel-L is an allogeneic, off-the-shelf cellular therapy product composed of mesenchymal stromal cells (MSCs) cryopreserved in a suspension of Plasma-Lyte A supplemented with human serum albumin (HSA) solution and 10% dimethyl sulfoxide (DMSO). The drug product (DP) is distributed in 6 mL cryovials with a label claim of 25 million cells per vial. The potency of each DP lot is measured by cell concentration, cell viability, and immunosuppressive activity as measured by an *in vitro* bioassay. The proposed indication is treatment of pediatric patients younger than 18 years of age with steroid-refractory graft-versus-host disease (SR-aGVHD), and the recommended treatment plan is a four-week course of twice weekly infusions of 2 million cells/kg.

5. **MAJOR MILESTONES**

Original BLA Submission Completed	January 31, 2020
Application Filed	March 30, 2020
Complete Response Letter Issued	September 30, 2020
BLA Resubmission Received	January 31, 2023
Resubmission Acknowledgement Letter Issued	March 7, 2023
Resubmission Internal Mid-Review Meeting	June 2, 2023
Cut-off Date for Resubmission Amendment Review	July 7, 2023
Resubmission PDUFA Action Date	August 2, 2023

6. CMC/QUALITY REVIEW TEAM

Reviewer/Affiliation	Section/Subject Matter
Matthew Klinker, PhD BLA Chair; CMC Reviewer OTP/OCTHT/DCT1/CTB2	CRL CMC Deficiency Comment #2 (Pre-license inspection) CRL CMC Deficiency Comment #3 (Potency assay) CRL Other CMC Comments 1-4, 5, 8
Heba Degheidy, MD, PhD CMC Reviewer OTP/OCTHT/DCT1/CTTB	CRL Other CMC Comment 6 (b) (4) assay validation)
Carolina Panico, MD, PhD CMC Reviewer OTP/OCTHT/DCT2/TEB2	CRL Other CMC Comments 7 (Extractable and leachable studies)

7. CONSULTS REQUESTED

Reviewer/Affiliation	Section/Topic	In agreement with consult recommendations?
Andrey Sarafanov OTP/OPPT/DH/HB2	CRL Other CMC Comments 7 (Extractable and leachable studies)	YES
Danielle Brooks, PhD OTP/OPT/DPT1/PTB3	CRL Other CMC Comments 7 (Extractable and leachable toxicology risk assessment)	YES

8. SUBMISSION(S) REVIEWED

Date Received	Submission	Sequence Number	Comments/Status
9/14/2020	125706/0.54	SN0054	Response to CMC RFI #15 and #31 (b) (4) assay validation)
1/31/2023	125706/0.65	SN0065	BLA Resubmission
3/9/2023	125706/0.68	SN0068	Response to CMC/DMPQ RFI #39 (inspection availability)
4/6/2023	125706/0.70	SN0070	Response to CMC RFI #42 (DP testing results and association with clinical outcomes)
4/27/2023	125706/0.74	SN0074	Response to CMC RFI #42 and Informal Teleconference on April 7, 2023 (DP testing results and association with clinical outcomes)
5/15/2023	125706/0.78	SN0078	Response to CMC RFI #47 (b) (4) assay validation and E&L studies)
5/22/2023	125706/0.80	SN0080	Proposed change to DCB retest potency acceptance criterion

Date Received	Submission	Sequence Number	Comments/Status
6/8/2023	125706/0.83	SN0083	Response to CMC RFI #51 (potency bioassay comparability and manufacturing capabilities)
6/22/2023	125706/0.85	SN0085	Response to CMC RFI #52 (potency bioassay comparability and manufacturing capabilities)
6/30/2023	125706/0.86	SN0086	Response to CMC RFI #52 (potency bioassay comparability)
6/30/2023	125706/0.88	SN0088	Response to CMC RFI #52 (potency bioassay comparability)

9. REFERENCED REGULATORY SUBMISSIONS

Submission	Holder	Referenced Item	Letter of Cross-Reference?	Comments/Status
MF5 (b) (4)	Lonza Walkersville, Inc	Manufacturing Facility for Lonza Walkersville (LWI)	Yes	Defer to DMPQ Reviewer
MF5 (b) (4)	Lonza Bioscience Singapore Pte, Ltd	Manufacturing Facility for Lonza Bioscience Singapore (LWI)	Yes	Defer to DMPQ Reviewer
MF2 (b) (4)	Mesoblast, Ltd	Manufacture of MSCs	N/A	Describes manufacturing process used to make remestemcel-L under IND
IND 7939	Mesoblast, Inc	Development History of remestemcel-L	N/A	IND under which clinical studies submitted with this application were conducted

10. REVIEWER SUMMARY AND RECOMMENDATION

A. EXECUTIVE SUMMARY

Background and Previous Actions

Remestemcel-L is a cryopreserved suspension of allogeneic culture-expanded MSCs derived from bone marrow aspirate collected from healthy human donors. Mesoblast, Inc. (“the Applicant”) submitted BLA 125706 in January 2020 requesting a license to market remestemcel-L in the US under the proprietary name RYONCIL for the treatment of SR-aGVHD in pediatric patients. In the initial BLA submission, the Applicant proposed two potency assays for lot release testing: (1) an (b) (4) for TNFR1 performed on MSC(b) (4), and (2) an *in vitro* bioassay measuring the capacity of each product lot to inhibit activation of allogeneic T cells (referred to as the inhibition of IL-2R α assay). CBER’s Office of Tissues and Advanced Therapies (OTAT) determined that these assays could not provide adequate control of the potency of the commercial product due to either poor performance characteristics (the inhibition of IL-2R α assay)

or the unknown relevance of the attribute measured to the therapeutic activity of the product (the TNFR1 assay). Additionally, OTAT found that the single-arm study submitted as the primary evidence of effectiveness (MSB-GVHD001, or “Study 001”) was not well-controlled and therefore did not meet the statutory requirement for substantial evidence of effectiveness. Because of these major clinical and CMC-related deficiencies, OTAT issued a complete response letter (CRL) denying approval of BLA 125706 on the action date (September 30, 2020) and recommended that the Applicant conduct at least one new clinical study to support the SR-aGVHD indication.

After receiving the CRL, the Applicant submitted a formal dispute resolution request (FDRR) in which they disputed OTAT’s position that Study 001 did not constitute substantial evidence of effectiveness (BLA 125706/0.60, SN0060, received April 1, 2021). This FDRR was accepted for review by CBER, and Dr. Peter Marks (Director, CBER) served as the designated reviewing official. In response to this FDRR, CBER argued that a meaningful potency assay was necessary to determine if the product administered in Study 001 was adequately “standardized as to [...] strength” as required for a trial to be considered well-controlled and adequate (21 CFR 314.126(d)). The Applicant conceded in the FDRR that the potency assay deficiency still needed to be addressed, and CBER deferred taking a position on whether Study 001 constituted substantial evidence of effectiveness until the Applicant had successfully addressed the potency assay deficiency. CBER indicated that they would consider the adequacy of the clinical data after the Applicant had addressed the related CMC issues and directed the Applicant to engage with OTAT regarding their planned approach to develop an acceptable potency assay.

Post-Action Interactions

As directed in the FDRR response, the Applicant provided updates on their progress resolving the CMC deficiency to OTAT during two interactions in 2021. In both updates, the Applicant acknowledged that the potency assays proposed in the original BLA submission were not suitable for controlling potency through routine lot release testing.

- The first update was provided in the briefing materials for a Type B meeting held under their IND (b) (4) to discuss a potential (b) (4)

A progress report was provided that described several potential potency assays in development, including new bioassays measuring the immunosuppressive activity of the product toward T cells and macrophages, and an assay measuring expression and/or activity of (b) (4) in the product.

The Applicant hypothesized that the same immunosuppressive activities of remestemcel-L thought to be the mechanism of action in treating SR-aGVHD would also mediate a therapeutic effect in (b) (4), and so these assays would also be relevant for this application. OTAT provided feedback indicating that the attributes measured by these assays appeared to be reasonable targets and recommended that the Applicant qualify their new potency assays before starting a new clinical study. During the meeting, the Applicant acknowledged that these methods were not yet suitable for routine lot

release testing and indicated that work on these assays was ongoing (meeting summary submitted in IND (b) (4))

- The second update was provided in briefing materials for a Type C meeting requested specifically to discuss the Applicant's plan for addressing the potency assay deficiency (BLA 125706/0.62, SN0063, received October 14, 2021). In this update, the Applicant indicated that the results of an investigation into the sources of variability for the inhibition of IL-2R α assay showed that this assay could not be salvaged, and proposed to use a newly developed bioassay measuring the immunosuppressive activity of the product toward T cells as the only potency assay for lot release. OTAT again agreed that this attribute was a reasonable target for a potency assay, but noted that a single bioassay may not be sufficient to address the potency assay deficiency as described in the CRL. OTAT also reiterated its position that at least one additional clinical study must be conducted before resubmission of BLA 125706 and recommended that the Applicant measure multiple product attributes during any new clinical trials to support potency assay development.

Resubmission Review

The Applicant has now submitted a complete response to the deficiencies described in the CRL (BLA 125706/0.65, SN0065, received January 31, 2023). None of the new assays discussed in previous interactions have been implemented, and the Applicant proposes to instead use a modified version of the inhibition of IL-2R α bioassay as the only potency test for lot release. This new version of the inhibition of IL-2R α bioassay (method C50826) has several differences from the previous version of this bioassay (method 21371) implemented to reduce variability. The most significant changes are:

(b) (4)

The 21371 version of the inhibition of IL-2R α assay was reviewed in the initial review cycle and found to have limited suitability as a potency test for lot release. The Applicant acknowledged these limitations during the original review cycle, stating that the 21371 assay "is currently not suitable as a quantitative assay" (BLA 125706/0.32, SN0032, Module 1.11.4, received June 15, 2020). The Applicant stated in a post-action interaction that they had conducted "an extensive investigation into the assay variability for the inhibition of IL-2R α assay" and concluded that "due to assay variance, the [inhibition of IL-2R α] assay is not suitable to be used as a critical quality attribute for DCB and DP batch release" (briefing materials in Module 1.6.1, BLA 125706/0.62, SN0063, received October 14, 2021).

The data provided in this resubmission demonstrate that the new C50826 assay does indeed have reduced variability relative to the previous 21371 assay. However, it was not initially clear how many DP lots had been tested with the new C50826 assay. DP batch release data provided in the initial resubmission reported potency in a single column labelled "IL-2R α inhibition" without distinguishing between the assay versions.

Through interactive review, the Applicant eventually provided a comprehensive dataset for all DP lots made using the proposed commercial process in which results from each assay version were reported separately (BLA 125706/0.74, SN0074; and BLA 125706/0.83, SN0083). This dataset showed that (b) (4) DP lots made during the ongoing commercial manufacturing campaign have been tested with the C50826 assay, but all DP lots made during the clinical manufacturing campaign in 2015-2016 (including all lots administered in Study 001) have been tested with the 21371 assay only. Therefore, the only potency data available for DP lots used in Study 001 was collected using the 21371 assay deemed unsuitable by the Applicant.

No data was provided in the resubmission to characterize how results from the C50826 assay compare to results from the 21371 assays, but the DP datasets provided through interactive review included results from (b) (4) commercial DP lots tested with validated versions of both the 21371 and C50826 assays. In analyzing the results from these (b) (4) commercial DP lots, we found no statistical relationship between the 21371 and C50826 assays by linear regression ($R^2 = (b) (4)$), and therefore conclude that potency as measured by the 21371 assay is not relevant to the C50826 assay.

To demonstrate that the attribute measured by the C50826 assay is related to the therapeutic effect of the product, the Applicant cites analyses purporting to link clinical outcomes to the average potency of DP lots received by each subject in Study 001. All potency data used in these analyses, however, was collected with the 21371 assay, so these analyses have no relevance to the C50826 assay. This failure to link the C50826 assay to clinical outcomes does not necessarily preclude approval, but the unestablished relevance of the attribute measured to the product's therapeutic effect may justify a more thorough approach that incorporates additional potency assays. The Applicant proposes an acceptance criterion of (b) (4) inhibition for the C50826 assay based on the potency of DP lots used in Study 001 as measured by the 21371 assay. However, this approach is not justified because potency measured by the 21371 assay is not reliably quantitative and has no statistical relationship to the C50826 assay. The Applicant therefore does not have an accurate measure of the potency of the product used in Study 001 with which to inform acceptance criteria for the C50826 assay. Additionally, all long-term stability studies provided by the Applicant measured potency using the 21371 assay. Given the low reliability of the 21371 assay and its lack of relationship to the C50826 assay, this stability data is not sufficient to demonstrate that the potency of the product will be stable over the Applicant's proposed 48-month shelf life.

After identifying these deficiencies, we engaged the Applicant several times during the review period to explain our position and provide an opportunity for them to address the issues. In SN0083, the Applicant argued that regression analyses comparing these assays should be (b) (4) (i.e., setting the (b) (4)), but the available data gives no indication that there is a relationship between these assays and therefore this approach is not justified. The Applicant also provided results from "comparative" studies in which material from (b) (4) DP vials was tested side-by-side with both C50826 and 21371 assays using the same (b) (4) lot for (b) (4) (SN0083; BLA 125706/0.85, SN0085, received June 22, 2023; and BLA 125706/0.86, SN0086, received June 30, 2023). However, the results from these studies did not convincingly

show a correlation between assays even under these best-case conditions. Additionally, these studies used modified versions of the C50826 assay rather than the validated C50826 assay, and so have little relevance to the validated C50826 assay. The Applicant's responses did not provide compelling evidence that potency measured by the 21371 assay has any relationship to potency as measured by the C50826 assay, and therefore did not adequately address these deficiencies.

Recommended Actions

We recommend that OTP deny the approval of this resubmission and issue a CRL due to the following deficiencies:

1. To ensure the continued potency of commercial product, an applicant should (1) identify product attributes linked to the therapeutic effect of the product, (2) develop tests to measure these attributes accurately and reliably, and (3) set acceptance criteria for these attributes in the commercial product to ensure consistency with product lots used in the efficacy studies supporting licensure. There is an acceptable scientific rationale supporting the Applicant's position that the attribute measured by the C50826 assay may be related to the therapeutic effect of the product, and the performance characteristics of the C50826 assay are generally acceptable for lot release testing purposes. But the C50826 assay fails to meet the third criterion because appropriate acceptance criteria cannot be determined in the absence of meaningful information on the potency of the product administered in Study 001. The Applicant has therefore not demonstrated that specifications of the approved product would "meet applicable requirements to ensure the continued [...] potency of such products" (21 CFR 601.2(d)).
2. Approved drug products must "bear an expiration date determined by appropriate stability testing" that includes "reliable, meaningful, and specific test methods" (21 CFR 211.137(a) and 21 CFR 211.166(a)(3)). The expiration date proposed by the Applicant is justified only by stability studies in which potency was measured by the 21371 assay, and the Applicant has acknowledged that this assay cannot be considered quantitative. The Applicant has therefore not conducted stability testing using a reliable or meaningful test for potency.

CBER previously indicated that the clinical data provided would be re-evaluated once the Applicant had resolved the potency assay deficiency and demonstrated that the product used in Study 001 was adequately "standardized as to [...] strength" as required for a trial to be considered well-controlled and adequate (21 CFR 314.126(d)). The Applicant failed to resolve the potency assay deficiency and the product lots administered in Study 001 remain poorly characterized. We therefore find that the Applicant has not met the requirements necessary for re-evaluation of the clinical data described in CBRE's response to the Applicant's FDRR but defer to the Clinical review team for evaluating the clinical data provided and determining if the Clinical deficiency has been adequately addressed.

B. RECOMMENDATION

I. COMPLETE RESPONSE

We have completed our review of all the submissions you have made relating to this BLA. After our complete review, we have concluded that we cannot grant final approval because of the deficiencies outlined below.

As stated in the Formal Dispute Resolution Request (FDRR) letter dated May 28, 2021, the Center for Biologics Evaluation and Research (CBER) was unable to determine that the product you used in Study MSB-GVHD001 was “standardized as to identity, strength, quality, purity, and dosage form” according to 21 CFR 314.126(d). Therefore, CBER was unable to confirm that the clinical data in the BLA provided substantial evidence of effectiveness of remestemcel-L for the treatment of SR-aGVHD in pediatric patients. This conclusion was based on insufficient information on the potency assay matrix as described in Comment #3 in the complete response letter (CRL) dated September 30, 2020.

In your BLA resubmission, you propose to implement a potency bioassay for lot release testing of remestemcel-L to address the deficiencies described in Comment #3 in the CRL. For the reasons explained in the comments below, we have determined that the data provided in your resubmission are not sufficient to demonstrate that your proposed potency bioassay and its acceptance criterion will ensure the continued potency of your product (21 CFR 601.2(d)). As a result, you have not resolved the deficiencies described in Comment #3 in the CRL, and therefore you have not demonstrated that the product used in Study MSB-GVHD001 was adequately standardized per 21 CFR 314.126(d) as requested in our FDRR response. The outstanding deficiencies are described in detail below.

Chemistry, Manufacturing, and Controls

1. In your resubmission (SN0065, dated January 30, 2023), you propose to control the potency of the remestemcel-L drug product (DP) using a bioassay measuring the inhibitory activity of your product toward T cells (referred to as the inhibition of IL-2R α assay). The bioassay you propose is a modified version of a similar bioassay that was reviewed during the original BLA review cycle and previously determined to not be suitable for release testing due to poor performance characteristics. The new version of this assay (C50826) incorporates several changes from the previous version (21371) intended to improve the assay's performance characteristics.

While we acknowledge that the changes implemented appear to improve assay performance, the data provided are not sufficient to demonstrate that the C50826 assay and proposed acceptance criterion provide meaningful control of the potency of remestemcel-L. For all DP lots used in the clinical study intended to provide the primary evidence of effectiveness (MSB-GVHD001), you tested for potency with the 21371 assay only with an acceptance criterion of (b) (4) inhibition. You use this same acceptance criterion for the C50826 assay. You have previously acknowledged, however, that the 21371 assay “is currently not suitable as a quantitative assay” (SN0032, Module 1.11.4, dated June 13, 2020), and we agree that the 21371 assay cannot be considered a reliable measure of product potency. Additionally, in our analysis of results for (b) (4) commercial DP lots

you provided during the review period (SN0074, dated April 26, 2023; and SN0083, dated June 8, 2023), we find no statistical relationship between the 21371 and C50826 assays by linear regression ($R^2=(b) (4)$). You provided results from your own analysis of these data in which you (b) (4)

(b) (4) but we do not agree that such an approach is justified. In the absence of an established statistical relationship between these assays, there is no justification for setting the acceptance criterion for the C50826 assay based on results from the 21371 assay. The C50826 assay and proposed acceptance criterion are therefore not sufficient to ensure the continued potency of the commercial product (21 CFR 601.2(d)) and therefore, you have not adequately addressed Comment #3 in the CRL dated September 30, 2020. To demonstrate that your potency assay(s) are appropriate for a licensed biologic, please provide data demonstrating that your potency assay(s) accurately and reliably measure a product attribute that is relevant to the intended therapeutic effect and have acceptance criteria that ensure that the potency of the commercial product is consistent with the potency of the product administered in the clinical trials submitted as evidence of effectiveness.

2. Establishing an assay that reliably measures product potency is necessary to establish a meaningful shelf life for your product. In Module 3.2.P.8.1 (SN0081, dated May 22, 2023), you provide a summary of completed and ongoing stability studies for the remestemcel-L DP. In Table 1, you indicate that Studies SP-011 and SP-013 are the primary long-term stability studies supporting your proposed 48-month shelf life for the cryopreserved remestemcel-L DP. You measured potency in SP-011 using the 21371 version of the inhibition of IL-2R α assay at all timepoints. SP-013 is ongoing, but the 21371 assay was used for all timepoints through 24 months, while the C50826 assay was used only for the most recent timepoint (36 months). As noted above in Comment #1, the 21371 assay is not suitable for use as a quantitative measure of potency and you have not adequately demonstrated a meaningful statistical relationship between the 21371 and C50826 assays. Studies SP-011 and SP-013 are therefore not sufficient to support your proposed shelf life. Expiration dates for approved DPs must be “determined by appropriate stability testing” that includes “reliable, meaningful, and specific test methods” (21 CFR 211.137(a) and 21 CFR 211.166(a)(3)).

Please provide data from stability studies that include at least one quantitative test for potency that measures a product attribute that is relevant to the intended therapeutic effect to support your proposed shelf life.

Labeling (from September 30, 2023 CRL)

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

Additional Non-CR Comments:

In addition to the deficiencies that were the basis for not granting approval, we have identified the following comments:

Chemistry, Manufacturing, and Controls

1. We completed a pre-license inspection of the Lonza Bioscience Singapore manufacturing facility in May 2023 and found no objectionable conditions. The second comment in our CRL (dated September 30, 2020) has therefore been resolved.
2. In Section 6.5 of your BLA resubmission (Module 1.11.4, SN0065), you address additional CMC comments from our CRL (dated September 30, 2020). After reviewing your responses to these comments, we have determined that additional CMC comments #1, 2, and 4-8 from the CRL have been adequately resolved or are no longer relevant due to new information provided in your resubmission and subsequent amendments.
3. In Section 6.5.3 of your BLA resubmission (Module 1.11.4, SN0065), you describe accelerated stability studies conducted to demonstrate that the inhibition of IL-2R α assay is stability indicating for (b) (4) (Study SP-019) and DP (Study SP-018) to address additional CMC comment #3 from the CRL (dated September 30, 2020). It is not clear, however, which version of the inhibition of IL-2R α assay was used in these studies. If you used the 21371 assay in the accelerated study for DP, we recommend that you repeat this study using the C50826 assay or other assays that can reliably measure product potency.
4. In Section 6.5.7 of your BLA resubmission (Module 1.11.4, SN0065), you describe a simulated-use extraction study you conducted for the DP container closure and a toxicological risk assessment of the compounds identified in this study. While this study is sufficient to support the use of the proposed DP container closure, leachable compounds from materials upstream in the manufacturing process also accumulate in the DP and may pose risk to those receiving your product. In general, for a BLA approval, we require assessment of total leachables presented in DP that accumulate throughout the manufacturing process, storage over the shelf-life and in-use hold. Such assessment should be performed in a real-time study using maximal hold times and temperatures at respective steps. In addition, this assessment (i) may use simulated intermediate solution(s) [without active ingredient and other complex compounds (e.g., cells, proteins) that may interfere with analytical detection of leachables], and (ii) may be started from a process step when no removal of potential leachables is performed (e.g., change of the buffer).

Therefore, please assess cumulative leachables from the upstream manufacturing steps. The study should evaluate leachables that accumulate after the (b) (4) to the final DP (e.g., from (b) (4) through final filling of the DP). In addition, we recommend that you include the assessment of leachables from upstream manufacturing steps in the design of the proposed protocol for the ongoing DP stability studies (PR-111), if feasible.

5. In Report RD-006 (Module 3.2.S.3.1, SN0065), you provided results from analyses purporting to show a relationship between product potency and clinical

outcomes in MSB-GVHD001. After we notified you of apparent discrepancies between the analysis methods described in this report and the clinical datasets used to conduct these analyses, you acknowledged that the analysis methods were "inaccurately described" in Report RD-006 and provided corrected results in SN0070 (dated April 5, 2023). Additionally, you provided further justification for analyzing potency data by grouping subjects and results from reverse cumulative distribution analyses in SN0074 (dated April 26, 2023).

We do not agree that the results reported in Report RD-006 and additional information provided in SN0070 and SN0074 establish an association between product potency and clinical outcomes because of limitations in the product potency dataset and analysis methods. The most significant of these limitations is that the analyses rely on potency data collected with the 21371 version of the inhibition of IL-2R α assay, and as noted above in Complete Response Comment #1, this assay has poor performance characteristics and no statistical relationship to the C50826 assay you proposed to use for lot release testing. Additionally, the moderate significance of the associations reported must be interpreted conservatively because of the large number of statistical tests performed and the post-hoc selection of analysis methods. Most subjects in MSB-GVHD001 received product from two or more DP lots and integrating potency from multiple lots into a single value for each subject limits the statistical power to detect a true relationship between product potency and clinical outcomes.

Establishing a relationship between product attributes and the product's clinical effect is not generally required for licensure. However, it will be very challenging for you to complete a convincing comparability exercise to support new DCB manufacturing without at least one product attribute with established relevance to the product's clinical efficacy. We therefore recommend that in future clinical studies you consider taking steps to avoid the limitations described above (e.g., pre-specify your analysis methods and ensure each subject receives product from only one DP lot) and thoroughly characterize the product used.

6. In SN0083 (dated June 8, 2023), you indicate that you can potentially treat (b) (4) patients with DP lots currently in inventory, and another (b) (4) patients with material from future DP lots made from your remaining DCB materials. In Section 6.3.5.1 of your BLA resubmission (Module 1.11.4, SN0065), you acknowledge that you will need to conduct a comparability exercise for new DCBs when the current stock is depleted. Additionally, you suggest that you will be able to demonstrate analytical comparability to support new DCBs using the inhibition of IL-2R α assay because the attribute this assay measures is associated with clinical outcomes.

As stated above in Additional Comment #5, however, we do not agree that you have demonstrated that the attribute measured by the inhibition of IL-2R α assay is associated with clinical outcomes, and therefore it is not clear that your current Critical Quality Attributes (CQAs) will be sufficient to demonstrate analytical comparability. Even if meaningful CQAs can be established, there is a reasonable likelihood that DP made from these new DCBs may not be comparable to DP made from the current DCB stock because your new DCB manufacturing process was developed more than 10 years after your current

DCBs were manufactured and new DCBs will be manufactured at a different facility (SN0085). As demonstrating analytical comparability to support new DCBs will therefore be very challenging, we recommend that you establish new DCB manufacturing and use DP made from these new DCBs in any new clinical studies.

7. A critical challenge for potency assays developed after completion of a clinical study is that the potency of DP lots used in the clinical study cannot be directly measured at the time of release, and data obtained by testing retained samples of these DP lots are difficult to interpret because the potency of the retained product may have degraded since release. As we have done previously (Preliminary Meeting Responses dated November 23, 2021), we again recommend that you qualify any new potency assays before initiating a new clinical study. This approach allows you to test product potency at the time of release and provides additional data to characterize the relationship between product attributes and clinical outcomes.

II. SIGNATURE BLOCK

Reviewer	Concurrence	Signature
Matthew Klinker, PhD Biologist OTP/OCTHT/DCT1/CTB2	Concur	Matthew W. Klinker -S Digitally signed by Matthew W. Klinker -S Date: 2023.07.31 10:56:18 -04'00'
Heba Degheidy, MD, PhD Staff Fellow OTP/OCTHT/DCT1/CTTB	Concur	Heba Degheidy -S Digitally signed by Heba Degheidy -S Date: 2023.07.31 11:01:33 -04'00'
Carolina Panico, MD, PhD Biologist OTP/OCTHT/DCT2/TEB2	Concur	Carolina Panico -S Digitally signed by Carolina Panico -S Date: 2023.07.31 11:27:59 -04'00'
Laura Ricles, PhD Chief, Tissue Engineering Branch 1 OTP/OCTHT/DCT2/TEB1	Concur	Laura M. Ricles -S Digitally signed by Laura M. Ricles -S Date: 2023.07.31 11:45:11 -04'00'
Melanie Eacho, PhD Director, Division of Cell Therapy 1 OTP/OCTHT/DCT1	Concur	Melanie Eacho -S Digitally signed by Melanie Eacho -S Date: 2023.08.01 08:26:54 -04'00'
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INTRODUCTION

A. Product Development Narrative

Osiris Therapeutics, Inc. (2002-2013)

Product development was initiated by Osiris Therapeutics, Inc. (OTI) under IND 7939.

(b) (4) in 2002 OTI began manufacturing an unrelated and non-HLA-matched allogeneic version of the product called “prochymal” that is the precursor of the proposed commercial product remestemcel-L. The OTI manufacturing process developed through several stages, and in 2007 manufacturing began under contract at the Lonza Walkersville (LWI) facility. At LWI manufacturing used a two-step process where MSCs were isolated from bone marrow aspirates (BMAs) and expanded through (b) (4) passages before cryopreservation as a drug substance intermediate called donor cell banks (DCBs), which are then thawed and further expanded through Passage (b) (4) and cryopreserved as the DP. This two-stage culture expansion is used throughout product development as well as in the proposed commercial manufacturing process.

OTI conducted two Phase 3 randomized, placebo-controlled studies in patients with acute graft-vs.-host disease (aGVHD): (1) Protocol 265 for adult patients in combination with corticosteroids, and (2) Protocol 280 for patients of all ages with steroid-refractory aGVHD (SR-aGVHD). In 2009 OTI began a rolling BLA submission (BLA 125334) while completing these studies, but both Protocol 265 and 280 failed to meet their primary endpoints and OTI withdrew the application before it was complete.

In anticipation of their licensing application, OTI applied to USAN for a proper name and prochymal was designated “remestemcel-L” by USAN in 2009. After the failure of their randomized control trials to meet primary endpoints, OTI maintained Protocol 275, an expanded-access protocol (EAP 275) under which pediatric patients with SR-aGVHD received remestemcel-L in addition to standard of care.

Mesoblast, Inc. (2013-present)

In 2013, the remestemcel-L development program was acquired by the Applicant, Mesoblast, Inc. (MSB). This acquisition included all remaining remestemcel-L DP and DCB material. MSB continued to treat subjects under EAP 275 using DP made by OTI, and reanalyzed data from the small number of pediatric patients (n=28) enrolled in OTI’s Protocol 280. Based on results from pediatric patients enrolled in 275 and 280, MSB proposed a single-arm study in pediatric patients to support licensure. This study (MSB-GVHD001, or “Study 001”) would use Day 28 overall response (OR) rate as the primary endpoint, and would be considered to have demonstrated efficacy if the results excluded a Day 28 OR rate of 45% as the null hypothesis.

To support Study 001, MSB proposed to manufacture new DP lots from DCB material acquired from OTI at Lonza’s Bioscience Singapore facility (LBSS). The Applicant was not able to demonstrate comparability between the new DP made at LBSS and OTI’s DP made at LWI, but FDA nonetheless agreed to allow MSB proceed with Study 001 using DP made at LBSS. A manufacturing campaign in 2015-2016 at the LBSS facility produced new DP lots from DCB material made by OTI. MSB then enrolled 55 pediatric

subjects with SR-aGVHD into Study 001, with all but 8 subjects receiving product made using the updated manufacturing process at LBSS. Study 001 met its primary endpoint (excluding an OR rate of 45% at Day 28).

MSB completed submission of BLA 125706 in January 2020 requesting a license to market remestemcel-L in the US for the treatment of SR-aGVHD in pediatric subjects with the results of Study 001 as the primary evidence of effectiveness. The application was accepted for filing and reviewed by CBER's Office of Tissues and Advanced Therapies (OTAT) with assistance from CBER Office of Compliance and Biologics Quality (OCBQ) and CDER's Oncology Center of Excellence (OCE). The application was not approved due to Clinical and CMC deficiencies, and on September 30, 2020, FDA issued a Complete Response Letter (CRL) to the Applicant.

B. Product and Manufacturing Process Overview

Remestemcel-L is a cryopreserved suspension of allogeneic culture-expanded MSCs derived from bone marrow aspirate collected from healthy human donors. The DP is formulated as described in the table below then stored and distributed in 6 mL vials containing 25×10^6 total cells in 3.8 mL

Table 1 - Remestemcel-L DP Formulation

Component	Concentration	Function
MSCs	(b) (4)	Drug Substance
25% Human Serum Albumin (HSA) Solution	20% v/v (5% w/v HSA)	Stabilization and protection of cells
Dimethyl Sulfoxide (DMSO)	10% v/v	Cryoprotectant
Plasma-Lyte A	70% v/v	Diluent providing physiological osmolarity and pH

Remestemcel-L is stored and transported in the vapor phase of liquid nitrogen at $\leq 135^\circ\text{C}$ until thawed for administration. Dosing is based on the patient's body weight at the time of infusion with a target dose of 2×10^6 MSCs/kg, and the product is administered intravenously by a qualified health professional.

The manufacturing process for remestemcel-L includes two distinct expansion phases:

1. The BMA is collected from healthy human donors, and MSCs are isolated from the BMA and expanded in culture through two passages. At the (b) (4) passage, MSCs are harvested and cryopreserved as a drug substance intermediate. Each lot of this drug substance intermediate is a DCB that is stored in (b) (4) which are subsequently thawed for further manufacturing use.
2. (b) (4) DCB is thawed and expanded in culture through three additional passages. At the (b) (4) passage, the cells are harvested, formulated, filled into cryovials, and cryopreserved as the DP.

The first step in manufacturing (production of the DCBs) was performed at LWI while under contract with OTI using a process that has since been retired, while the second

step in manufacturing is ongoing at LBSS and uses DCB material made at LWI in 2008-2009.

Third-party testing laboratories performed most lot release testing for remestemcel-L DP on samples from filled DP vials. The table below summarizes DP specifications.

Table 2 - Remestemcel-L Specifications

Attribute	Assay	Specification	Sample	Testing Facility
In-Process Sterility	(b) (4)	Negative	(b) (4)	(b) (4)
Mycoplasma	(b) (4)	Negative	(b) (4)	(b) (4)
Sterility	(b) (4)	Negative	Filled DP Vial	(b) (4)
Purity	Endotoxin (b) (4)	(b) (4)	Filled DP Vial	(b) (4)
Identity	(b) (4)	(b) (4) CD166 ⁺	Filled DP Vial	(b) (4)
Identity	(b) (4)	(b) (4) CD105 ⁺	Filled DP Vial	(b) (4)
Identity	(b) (4)	(b) (4) CD45 ⁺	Filled DP Vial	(b) (4)
Potency	IL-2R α Inhibition	(b) (4) Inhibition of IL-2R α in (b) (4)	Filled DP Vial	(b) (4)
Potency	Cell Viability	(b) (4) Viability	Filled DP Vial	(b) (4)
Potency	Cell Concentration	$\geq 6.68 \times 10^6$ cells/mL	Filled DP Vial	(b) (4)
Appearance	(b) (4)	(b) (4)	Filled DP Vial	(b) (4)
Purity	Residual BSA (b) (4)	(b) (4)	Filled DP Vial	(b) (4)
Purity	Residual Trypsin	(b) (4)	Filled DP Vial	(b) (4)
Appearance	Visual Inspection and (b) (4) Sampling	(b) (4)	Filled DP Vial	LBSS

C. Original BLA Submission Review Summary

Major clinical and CMC deficiencies identified during the original BLA review cycle are summarized below.

Clinical Deficiency: A single-arm study (MSB-GVHD001, or “Study 001”) was the primary evidence of effectiveness provided in the application. The results of Study 001 were compared to historical controls and this study achieved its primary endpoint by excluding an OR on Day 28 of 45% (OR 69.1%; 95% CI 55.2%-80.9%). Although some

members of the clinical review team recommended approval based on these results, the study design was ultimately determined to be prone to bias and therefore Study 001 was not considered an adequate and well-controlled study. OTAT recommended that the Applicant complete at least one randomized clinical trial in a similar patient population.

CMC Deficiency: The presumed mechanism of action for remestemcel-L is immunomodulatory activity and *in vitro* assays reliably show that remestemcel-L can inhibit T cell activation, but the extent of this inhibition is variable and difficult to measure. At the time of original submission of this application, two assays for product potency were in place: (1) an (b) (4) for TNFR1 performed on MSC (b) (4), and (2) an *in vitro* bioassay intended to measure the capacity of each product lot to inhibit activation of allogeneic T cells (referred to as the inhibition of IL-2R α assay). We engaged the Applicant during the original review cycle regarding several deficiencies for these assays, and by the end of the review cycle we determined that these assays were not acceptable as potency assays for the following reasons:

- The basis for selecting TNFR1 as an attribute related to potency was a series of experiments conducted in 2005 using a previous version of the remestemcel-L product. These initial experiments showed that knockdown of TNFR1 reduced the capacity of MSCs in the precursor product to inhibit T cell proliferation (Report R-045-05). During the original review cycle, the Applicant committed to repeating these experiments using the proposed commercial product. The results from these new experiments showed that “the immunomodulatory effects of remestemcel-L on activated T cell proliferation *in vitro* are independent of TNFR1 activity and expression,” directly contradicting the stated rationale supporting a relationship between this product attribute and product potency. The Applicant also provided analyses of TNFR1 levels with clinical outcomes, and results from experiments intended to show a relationship between TNFR1 levels and immunomodulation of human monocytes, but both approaches had significant limitations and did not support the Applicant’s conclusions. We determined that the Applicant had not demonstrated that TNFR1 levels had any relevance to the activity of the product, and therefore had not provided adequate justification for considering this an assay for product potency.
- During review of the application, we asked the Applicant to address an apparent reduced potency of lots made in 2019-2020 for initial commercial release relative to lots used in Study 001 as measured by the inhibition of IL-2R α assay. The Applicant attributed this difference to variability in the assay and indicated that the assay was too variable to be used as a quantitative assay for lot release. They proposed to reclassify this as a qualitative assay for “activity” rather than an assay for potency.

As the TNFR1 assay was determined to measure an attribute of unknown relevance, and the inhibition of IL-2R α assay performed too poorly to be considered a quantitative assay for lot release, the CMC review team recommended that this product not be approved because the Applicant did not have an acceptable potency assay. In the CRL, the Applicant was asked to provide data demonstrating that product attributes measured by potency assays used for lot release and establishing stability have a statistically

meaningful relationship to clinical outcomes, surrogate markers of *in vivo* activity, or a relevant product activity as measured by an *in vitro* biological assay.

D. Post-CRL Interactions

1. Post-Action Type A Meeting – Teleconference on November 17, 2020 (125706, Amendment 56)

Shortly after the CRL was issued, the Applicant requested a Type A meeting to discuss the decision to not approve the BLA. The Applicant proposed that an accelerated approval be granted with a post-approval study of SR-aGVHD in subjects >12 years of age to be completed as part of the accelerated approval agreement. This proposal included some unspecified improvements to the inhibition of IL-2R α assay, and PMCs for the development of assays related to (b) (4) expression/activity and TNF α -induced secretion of MCP-1/CCL2 and M-CSF, which are mediators the Applicant speculated were important for the activity of the product toward macrophages/monocytes.


OTAT did not agree with the accelerated approval proposal and again asked the Applicant to complete at least one randomized trial to support licensure. The CMC review team provided some feedback on the Applicant's proposed new potency assays, noting that they appeared to measure reasonable targets, but the adequacy of the proposed potency matrix could not be determined at this time. Additionally, we recommended that all new potency assays be qualified before initiation of their new clinical trial and agreed to provide guidance on assay development during later interactions.

2. Formal Dispute Resolution Request – Decision letter issued to Applicant on May 28, 2021

The Applicant appealed OTAT's decision regarding the adequacy of their single-arm trial in a formal dispute resolution request (FDRR), with CBER director Peter Marks assigned as the designated reviewing official. The Applicant asked for reconsideration "on the narrow issue of whether the clinical data contained in the BLA provide substantial evidence of effectiveness," and acknowledged that, even if this appeal were successful, they would still need to address the potency assay deficiency. CBER took the position that the lack of a meaningful potency assay meant that it could not be established that the product used in the single-arm study was "standardized as to identity, strength, quality, purity, and dosage form" as required for a clinical study to be considered "adequate and well-controlled" [21 CFR 314.126(d)]. On these grounds, CBER deferred reconsideration of the available clinical data until the potency assay deficiency has been addressed and indicated in response to the FDRR that "CBER would be happy to consider the adequacy of the clinical data when you have addressed the related CMC issues."

3. (b) (4) Meeting – Teleconference on (b) (4) (IND (b) (4), Amendment 28)

(b) (4)



4. Type C Meeting – Teleconference on November 29, 2021

The Applicant requested a Type C meeting in late 2021 to discuss their plan for addressing the potency assay deficiency (BLA 125706, SN0063, received October 14, 2021). In the briefing material for this meeting, the Applicant indicated that both potency assays from their original BLA submission would be used “for remestemcel-L characterization purposes only.” Additionally, the Applicant reported that they “undertook an extensive investigation into the assay variability for the inhibition of IL-2R α assay” and concluded that “due to assay variance, the [inhibition of IL-2R α] assay is not suitable to be used as a critical quality attribute for DCB and DP batch release” (SN0063, briefing materials in Module 1.6.1, received October 14, 2021). They proposed to use a new bioassay (referred to as the (b) (4) assay, or (b) (4)) as the only potency tests for lot release in their resubmission. The CMC team again agreed that this attribute was a reasonable target for a potency assay but noted that a relationship between this attribute and the product’s clinical effect had not established and that a single bioassay may not be sufficient to address the potency assay deficiency as described in the CRL. OTAT also reiterated its position that at least one additional clinical study must be conducted before resubmission of BLA 125706 and recommended that the Applicant measure multiple product attributes during any new clinical trials to support potency assay development.

E. Review Memorandum Organization

The Applicant provided a complete response document in Module 1.11.4 (SN0065, received January 31, 2023) that provides a point-by-point response to all comments in the CRL. The review below follows this same organization and will address the Applicant's responses to the CRL in two sections:

- A. Complete Response Deficiencies: The CRL included 4 comments describing the deficiencies that prevented approval.
- B. Additional CMC Deficiencies: Eight (8) additional CMC comments were included in the CRL describing unresolved CMC deficiencies that were not part of the basis for not approving the application and providing recommendations and advice.

RESUBMISSION REVIEW

A. Complete Response Deficiencies

1. Clinical Deficiency (CRL Comment #1)

Overall Assessment Response to CRL Comment #1:

We defer to the Clinical review team on the adequacy of the response provided.

2. Pre-license Inspection (CRL Comment #2)

A pre-license inspection was conducted by inspectors from OTP and OCBQ/DMPQ May 10-19, 2023. No objectionable conditions were observed.

Overall Assessment Response to CRL Comment #2:

The pre-license inspection was completed, and no objectionable conditions were found. Both DMPQ and OTP/OCTHT reviewers find the Lonza Singapore facility acceptable for commercial manufacturing.

CRL Comment #2 has been resolved. See non-CR Comment #1.

3. Potency Assay Deficiency (CRL Comment #3)

CRL Comment #3 asked the Applicant to:

1. "Identify all assays that you consider tests for product potency and provide justification for how controlling the attributes measured by these assays is adequate to ensure that each lot of remestemcel-L has acceptable levels of product activity."
2. "Provide data demonstrating that the product attributes measured by potency assays used for lot release and establishing stability have a statistically meaningful relationship to clinical outcomes, surrogate markers of *in vivo* activity, or a relevant product activity as measured by an *in vitro* biological assay."

In this resubmission, the Applicant proposes to use a modified version of the inhibition of IL-2R α bioassay as the only potency test for lot release. This new version of this bioassay (method C50826) differs from the previous version of this bioassay (method 21371) in several important ways that are intended to reduce assay variability: ^{(b) (4)}

To support the new C50826 bioassay, the Applicant provided the following data in the initial resubmission amendment (SN0065):

- (b) (4) [Redacted]

[Redacted]

[Redacted]

[Redacted]

[Redacted]

DP Potency Results for 21371 and C50826 Assays

(b) (4) [Redacted]

Conclusions

(b) (4)

DP Stability and Proposed Shelf Life

The Applicant proposes a 48-month shelf life for cryopreserved DP, justified by results from primary long-term stability studies SP-011 and SP-013 (Table 1, Module 3.2.P.8.1, SN0081). Study SP-011 completed testing through 48 months, but potency was measured exclusively for the 21371 assay (Table 2, Module 3.2.P.8.1, SN0081). Study SP-013 is ongoing with results through 36 months currently available, but the C50826 potency assay was used only at the 36-month timepoint (Table 4, Module 3.2.P.8.3, SN0065). Additionally, potency for 2 of the (b) (4) DP lots measured by the C50826 assay at 36 months was below the proposed (b) (4) inhibition acceptance criterion (b) (4) inhibition for lot (b) (4), and (b) (4) inhibition for lot (b) (4), with another barely passing (b) (4) inhibition for lot (b) (4).

Reviewer Comments:

- The Applicant has acknowledged that the 21371 assay is not suitable as a quantitative test, and a qualitative assay has limited value in studies attempting to establish stability of the product. Results from SP-011 therefore are not sufficient to support a proposed shelf life because potency in this study was tested exclusively with the 21371 assay.

- In Study SP-013, the C50826 assay was used only for the most recent timepoint (36 months) and potency at all previous timepoints was measured using the 21371 assay. Without reliable potency data for these DP lots at the time of release, however, these 36-month C50826 results cannot be interpreted.

The Applicant therefore cannot set a meaningful expiration time for their product because they have not used an appropriate quantitative potency assay in their stability studies.

Overall Assessment Response to CRL Comment #3:

Major Deficiencies:

- The C50826 bioassay proposed as the sole potency test for lot release cannot ensure the continued potency of the commercial product because there is no reliable potency data for the DP lots administered in Study 001. While the C50826 assay may have better performance characteristics than the previous 21371 version, it cannot provide meaningful control of product potency without appropriate acceptance criteria. See CRL Comment #1.
- The 21371 potency assay used in stability studies is not suitable as a quantitative assay, and the stability data provided therefore cannot establish a meaningful expiration time for this product. See CRL Comment #2.

Minor Deficiency:

- The analyses provided by the Applicant purporting to show a link between product potency and clinical outcomes are limited by the use of data from the 21371 assay, the post-hoc selection of analysis methods, and the fact that most subjects received product from multiple lots. While a potency-related CQA link to clinical outcomes may not be strictly required for approval of this application, the Applicant should attempt to find such CQAs in future clinical studies and take steps to avoid the limitations described above. See non-CR Comment #5.

4. Labelling (CRL Comment #4)

Overall Assessment of Response to CRL Comment #4:

Labeling review was not completed in this review cycle due to deficiencies preventing approval.

Major Deficiency: CRL Comment #4 has not been resolved, and we defer comment on the proposed labeling until the application is otherwise acceptable for approval. See CRL Comment #3.

Additional CMC Deficiencies

1. Lot Sampling Approach

“[...] We therefore recommend that you develop a more thorough lot sampling approach that includes testing multiple vials from each lot, and that you revise your specifications for [your potency] assays to account for the distribution of values obtained from multiple vials within a lot. For each assay, you should choose a minimum acceptable result for each vial, then perform a statistical analysis to determine how to set your specifications to ensure that the frequency of vials below this minimum value is acceptably low.”

In Report MR-186, the Applicant describes studies conducted to validate the C50826 version of the inhibition of IL-2R α bioassay. (b) (4)

[REDACTED]

[REDACTED]

(b) (4)

(b) (4)

(b) (4)

[REDACTED]

Reviewer Comments:

- (b) (4)

[REDACTED]

[REDACTED]

[REDACTED]

Overall Assessment of Response to Additional CMC Comment #1:

While we do not agree with using potency data collected with the 21371 assay to set acceptance criteria for the C50826 assay, the Applicant's approach to developing a multiple-vial testing strategy for the C50826 assay appears to be reasonable. This comment can be considered resolved with regard to the multiple-vial testing approach, but this issue can be revisited if new potency assays are introduced in subsequent review cycles.

Additional CMC Comment #1 has been resolved. See non-CR Comment #2.


2. Improvements to Inhibition of IL-2R α Assay

"[...] If you intend to continue using [the inhibition of IL-2R α] assay for DP lot release, we recommend that you continue to develop this assay and revise the testing procedure as appropriate to improve robustness to (b) (4) variability and provide more consistent results."

Although the Applicant's position in 2021 was that the inhibition of IL-2R α assay could not be improved enough to make it acceptable for lot release testing purposes, a revised version of this assay is now proposed to be the only release test for potency.

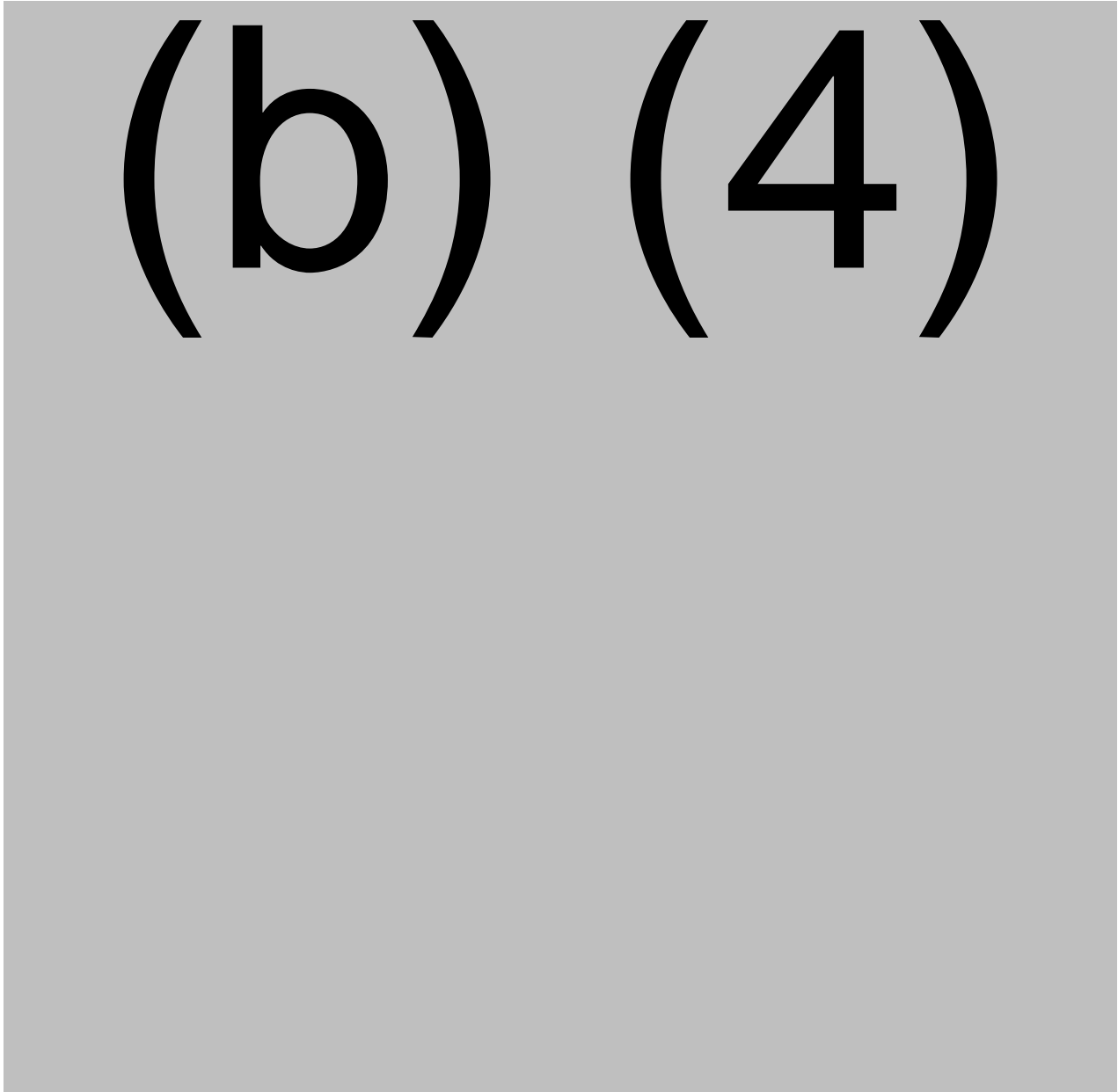
This new version of the inhibition of IL-2R α bioassay (method C50826) has several differences from the previous version of this bioassay (method 21371) intended to reduce variability. The most significant changes are: (b) (4)

(b) (4)

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Validation of the C50826 assay was described in Report MR-186 (“Summary Report: Assay Validation of the Determination of IL-2R α Inhibition on (b) (4) by ceMSCs”). The table below summarizes these validation studies and their acceptance criteria.

(b) (4)

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(b) (4)

Overall Assessment of Response to Additional CMC Comment #2:

The C50826 version of the inhibition of IL-2R α bioassay appears to have better performance characteristics relative to its predecessor 21371 version, and variability in potency as measured by the C50826 assay is reduced further by (b) (4) potency results from (b) (4) test samples. The C50826 assay can be considered quantitative, but other limitations (lack of data for DP lots, unknown relationship to clinical outcomes) make it unclear if this assay alone would be sufficient to control potency of the product.

The Applicant has followed our recommendation so Additional CMC Comment #2 can be considered resolved, but limitations of this assay beyond its performance characteristics make it unsuitable as a lot release potency assay. See non-CR Comment #2.

3. Stability-Indicating Assays for DCB Testing

"In the Chemistry, Manufacturing, and Controls Information Request #23, we state that assays established as stability-indicating for the DP may not be stability-indicating for the DCB and recommend that you establish these assays as stability-indicating for the DCBs in addition to the DP. In Amendment 34 (dated June 18, 2020) (b) (4)

Please note that assays that are not established as stability-indicating specifically for DCB material may not be relevant for use in future comparability exercises performed after changes are made to the DCB manufacturing process."

(b) (4)

. While this is reasonable position, data should still be provided to confirm this assumption.

The resubmission included data from a new accelerated stability study in which material (b) (4) -

(b) (4)

[Redacted]

[Redacted]

[Redacted]

- [Redacted]
- [Redacted]
- [Redacted]

[Redacted]

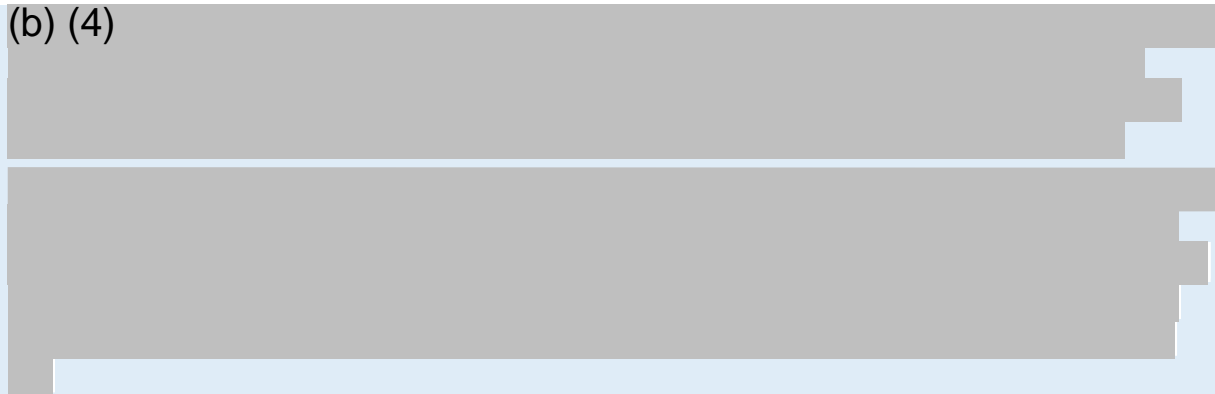
[Redacted]

- [Redacted]
- [Redacted]

Overall Assessment of Response to Additional CMC Comment #3:


The Applicant has now provided results from accelerated stability studies to demonstrate that (b) (4)

(b) (4)

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4. *Extractable Volume Justification*

(b) (4)

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Overall Assessment of Response to Additional CMC Comment #4:

The Applicant provided additional justification in briefing materials for a post-action Type A meeting in 2020 (SN0056, received October 26, 2023), and we found this justification to be acceptable (see FDA's preliminary responses in Module 1.6.3, SN0060, received April 1, 2023).

Additional CMC Comment #4 has been resolved. See non-CR Comment #2.

5. (b) (4) Testing (b) (4) Lot Variability

“During validation of the (b) (4) for TNFR1 and IL-2Rα that are used for DP lot release testing, you evaluated the effects of (b) (4) different lots of each (b) (4) however, this evaluation was performed using results obtained from different DP lots. We recommend that you characterize variability in performance between

lots of (b) (4) by using (b) (4) lots to test the same test article (i.e., the same cell (b) (4)).”

The TNFR1 assay will no longer be used for lot release testing, so no additional information is necessary for this assay.

For the inhibition of IL-2R α assay, the Applicant designed their validation of the new C50826 version of this assay to address this recommendation. In these validation studies, (b) (4) tested with (b) (4) IL-2R α (b) (4) and the results met the pre-specified acceptance criterion of %RSD (b) (4) (%RSD of (b) (4) test runs was (b) (4)).

Overall Assessment of Response to Additional CMC Comment #5:

The Applicant followed this recommendation, and the results met the prespecified acceptance criterion, so this response is acceptable.

Additional CMC Comment #5 has been resolved. See non-CR Comment #2.

6. (b) (4) Assay Validation

(b) (4)

(b) (4)

(b) (4)

(b) (4)

(b) (4)

Overall Assessment of Response to Additional CMC Comment #6:

The functional range over which linearity was assessed is suitable for each assay, and no additional validation studies are necessary. The assays appear to be robust to changes in instrument and reagent lot. The revised test record for the Applicant's (b) (4) assays appears to be adequate.

Additional CMC Comment #6 has been resolved. See non-CR Comment #2.

7. Extractable and Leachable Studies for DP Container Closure

"In Amendment 45 (dated July 21, 2020), you commit to submitting results from additional extractable and leachable studies to support the use of the 6 mL (b) (4) Vials as container closure for the DP. As these reports were not submitted before the action date for this application, they were not reviewed, and additional information may be needed after the results are reviewed."

During the original review cycle, extractable and leachable studies conducted for DCB and DP container closures were reviewed by consult reviewers from CDRH. They identified several deficiencies in the studies provided and we asked the Applicant to address these deficiencies in RFI #32. In the Applicant's response (Module 1.11.1, SN0045, received July 21, 2020), they proposed to conduct additional studies in a (b) (4) phase testing approach, but all studies proposed would not be completed by the action date. The Applicant requested feedback on the studies proposed in the briefing document for the post-action Type A meeting (SN0056, received October 26, 2023), and we provided recommendations and acknowledged that the proposed studies appeared to be acceptable in our responses (see meeting summary in Module 1.6.3, SN0060, received April 1, 2023).

In their complete response, the Applicant indicates that they conducted a simulated use extraction rather than the exaggerated extraction proposed in SN0045. Included in the resubmission were a report describing the compounds identified (Report V1NNF520, Module 3.2.P.2), and a toxicological risk assessment for the compounds identified (Module 3.2.P.2), and protocol for evaluating leachables from the container closure in ongoing DP stability studies. (PR-111, Module 1.11.4). We obtained consult reviews from Dr. Andrey Sarafanov (CBER/OTP/OPPT/DH/HB2) for the extractable study, and from Dr. Danielle Brooks (CBER/OTP/OPT/DPT1/PTB3) for the toxicological risk assessment. Their conclusions are briefly summarized below.

Evaluation of Simulated Use Extraction Study for DP Container Closure

We provided the report for the simulated use extraction study and the protocol proposed for the evaluation of leachables in the ongoing stability studies, as well as the relevant section of the Applicant's complete response document (Section 6.5.7.1, Module 1.11.4) to Dr. Sarafanov for review. For additional context, we also provided the accompanying toxicological risk assessment for the extractable study, the Applicant's description of the DP container closure (SN0003, Module 3.2.P.7), and the Applicant's previous

toxicological risk assessment provided during the original review cycle (Report C20453.2, Module 3.2.P.2, SN0039, received July 2, 2020).

Dr. Sarafanov determined that the results from the simulated use extraction study appeared acceptable but noted that the Applicant did not specify the amount of each organic leachable compound found in this study, only that the amounts found were below the analytical evaluation thresholds. We therefore asked the Applicant to provide this information in RFI #47, and the Applicant did so in SN0078 (received May 15, 2023). Dr. Sarafanov found this response to be adequate and concluded that the information provided for assessment of leachable from the DP container closure was acceptable.

Although the DP container closure studies and the protocol proposed for the assessment of the leachables from the container closure in the ongoing DP stability studies were determined to be acceptable, Dr. Sarafanov noted that the Applicant had not conducted studies to assess leachable compounds that may accumulate in the DP from upstream processes. Dr. Sarafanov therefore recommends that the Applicant complete a study to evaluate cumulative leachable compounds that includes manufacturing process steps from (b) (4) through final filling. Dr. Sarafanov indicated that an assessment of cumulative leachable compounds in the DP is generally required for approved products per CBER policy, but that such a study can likely be conducted as a post-marketing commitment. For more details on this review, please refer to Dr. Sarafanov's review memorandum in CBER Connect (STN_125706-65_Mesoblast_Remestemcel_BLA_CMC-Review Memo_2023-06-09.pdf).

Toxicological Risk Assessment of Leachable Compounds for DP Container Closure

We provided the toxicological risk assessments for the leachable study provided in the resubmission (SN0065) and a risk assessment for extractable/leachable studies submitted during the original review cycle (Report C20543, SN0039) to Dr. Brooks for review. Dr. Brooks concluded that the risk assessments provided were acceptable and supported the use of the Applicant's proposed DP container closure system. For more details on this review, please refer to Dr. Brooks' review memorandum in CBER Connect (125706.000_EandL_TRA.pdf).

Overall Assessment of Response to Additional CMC Comment #7:

This comment has been resolved as the studies provided for the DP container closure are acceptable. However, a non-CR comment will be provided to the Applicant to address cumulative leachable compounds in their next resubmission.

Minor Deficiency: Please assess potential leachable compounds in upstream manufacturing steps occurring after the (b) (4). See non-CR Comment #4.

8. DP Stability and Sterility Sample Shipping Validation

“In your response to FDA late-cycle meeting materials (Amendment 52, dated July 23, 2020), you commit to providing an updated assessment of DP stability using the agreed-upon revised DP specifications, and a final study report supporting microorganism recovery in release and in-process sterility samples shipped under various conditions to the (b) (4) for testing. You provided your responses in Amendment 55 (dated September 18, 2020); however, this amendment was not reviewed due to receipt late in the review cycle. You also committed to providing a written plan for the periodic endotoxin testing of incoming lots of product contact materials; however, this information was not received at the time of this letter issuance. Additional information may be requested after review of these materials.”

The Applicant submitted a reassessment of DP stability due to specification changes and data supporting hold times and shipping conditions for in-process and DP sterility testing samples in SN0055 (received September 21, 2020). As this amendment was received after the amendment review cut-off date, however, it was not reviewed during the original review cycle. Additionally, the Applicant agreed to submit a plan for testing incoming product-contact materials for endotoxin following the Late-Cycle meeting during the original review cycle (see response to meeting discussion in Module 1.11.1, SN0052, received September 2, 2020).

Reviewer Comments:

- The Applicant's complete response document does not address the reassessment of DP stability they provided in SN0055 and focuses only on addressing the suitability of the storage/shipping conditions used for sterility samples and testing of incoming materials for endotoxin. For review of DP stability and the Applicant's proposed DP shelf life, please see Complete Response Deficiency #3 above.
- Although the deficiencies regarding storage/shipping conditions for sterility samples and endotoxin testing of incoming materials were raised by the DMPQ reviewer during the original review cycle, we will review the Applicant's response to these deficiencies here.

Hold Times for Reagent and In-Process Sterility Testing Samples

Sterility testing of in-process samples is now conducted primarily by a third-party testing laboratory in (b) (4), but these samples were shipped to a testing facility in the (b) (4) in the original review cycle. In Report SGTS-26625 (Module 1.11.4, SN0065), the Applicant summarizes a study conducted by LBSS personnel to establish hold times for in-process and reagent samples intended for sterility testing. This study used (b) (4) test organisms as well as (b) (4) environmental isolates from their manufacturing facility (b) (4). Test articles were (b) (4).

The table below summarizes the test samples and maximum hold times as determined by this study.

Table 7 - Hold Times for Remestemcel-L Reagent and In-Process Sterility Samples

(b) (4)

Reviewer Comments:

- The data provided support the hold times proposed for reagent and in-process sterility samples. Additionally, the Applicant now uses a facility in (b) (4) for sterility testing, so samples will no longer be shipped to the testing facility in the (b) (4) unless the (b) (4) facility is not available. Hold times for sterility samples are therefore will in most cases be much (b) (4).
- Similar studies were conducted for DP sterility samples during the original review cycle and determined to be adequate.

Endotoxin Testing for Incoming Materials as LBSS

The Applicant indicates that the primary DP container closure (6 mL vial from (b) (4)) is tested for endotoxin by the manufacturer. Personnel at LBSS tested (b) (4) unique lots of these containers for endotoxin and will continue to test (b) (4) (b) (4). All other product-contact consumables are purchased from vendors qualified by LBSS and are either FDA-cleared medical devices or tested for endotoxin by the manufacturer. All incoming material are subject to receiving activities that include verifying that endotoxin testing is included on material certificates of analysis.

Reviewer Comment: The Applicant's approach to ensuring that incoming materials are relatively free from endotoxin appears to be reasonable. Receiving activities and associated SOPs were reviewed during inspection of the LBSS facility by OTP and DMPQ reviewers and were found to be acceptable.

Overall Assessment of Response to Additional CMC Comment #8:

The data provided support the Applicant's proposed hold times for reagent and in-process sterility samples, and the approach to ensuring that product-contact materials are relatively free from endotoxin is acceptable.

Additional CMC Comment #8 is resolved. See non-CR Comment #2.