

## DBSQC/OCBQ ANALYTICAL METHOD REVIEW MEMO

**To:** The file STN 125755/0

**From:**

Reviewer	Role	Date finalized	Stamp	Supervisor	Stamp
Seth Schulte	Reviewer	9/8/2022		James Kenney	
Tao Pan	Reviewer	06/09/2022		Tao Pan	
Nahid M. Parvin	Reviewer	08/26/2022		Muhammad Shahabuddin	
Esmeralda Alvarado Facundo	Lead Reviewer	8/30/2022			

**Through** Maryna Eichelberger, Ph.D.  
Division Director, DBSQC/OCBQ

**Applicant:** bluebird bio, Inc

**Subject:** Review of Analytical Methods used for Lot Release of Skysona (elivaldogene autotemcel) Lenti-D Lentiviral Vector (LVV), Drug Substance (DS) and Drug Product (DP).

**Recommendation:** Approval with PMC agreed upon.

### Executive Summary:

The following analytical methods used for lot release of Skysona and the associated method validations or qualifications, were reviewed:

1. Sterility test of (b) (4) DP (Seth Schulte)
2. Endotoxin test of (b) (4) DP (Seth Schulte)
3. (b) (4) test of (b) (4) DP (Seth Schulte)
4. (b) (4) (Tao Pan)
5. (b) (4) (Tao Pan)
6. Appearance of (b) (4) DP (Tao Pan)
7. (b) (4) (Nahid M. Parvin)
8. (b) (4) (Esmeralda Alvarado Facundo)
9. (b) (4) (Esmeralda Alvarado Facundo)

**Conclusion:**

The analytical methods and their validations and/or qualifications reviewed for the Skysona LVV and/or drug product were found to be adequate for their intended use, except for the outstanding issues for testing (b) (4) of (b) (4) drug product, for which bluebird bio has provided written commitment to resolve as Postmarketing Commitments.

**Documents Reviewed:**

Information in sections of the original submission that describe control of LVV, DS and DP (3.2.S.4 and 3.2.P.5, respectively), including descriptions of LVV, DS and DP specifications, analytical procedures of LVV, DS and DP and validation of these analytical procedures were reviewed. Additional information in amendments specified by each reviewer were also reviewed.

**Background:**

bluebird bio, Inc submitted BLA STN 125755 on October 18, 2021, for Skysona (eli-cel or elivaldogene autotemcel), which is a gene therapy. Skysona consists of autologous CD34+ cell enriched population of Hematopoietic Stem Cell (HSC) transduced ex-vivo with Lenti-D LVV which contains human adenosine triphosphate (ATP)-binding cassette subfamily D, member 1 (ABCD1) gene that encodes a functional human adrenoleukodystrophy protein (ALDP). The Lenti-D LVV is generated by transient transfection of human embryonic kidney (HEK293T) cells, and it contains structural proteins, a lipid envelope and two copies of (b) (4)-strand RNA that encodes ABCD1 and genetic elements necessary for the LVV function. After transfection, the LVV is harvested, purified and filled into a (b) (4) vial and stored at -65 °C. The Lenti-D LVV is used to transduce the HSC, after transduction cells are washed (b) (4) Skysona (b) (4). To produce the Skysona DP, the DS is formulated in (b) (4) cryopreservation solution containing 5% dimethyl sulfoxide (DMSO) and stored at (b) (4) in vapor-phase liquid nitrogen.

Due to limited availability of patient samples, some DP testing was performed using healthy donor CD34+ cells transduced with either Lenti-D LVV or BB305 LVV (Zynteglo, STN 125717), a similar product manufactured by bluebird bio.

DBSQC reviews BLA and their supplements to ensure the analytical methods are appropriately described, validated and suitable for intended purposes. The following analytical methods used for LVV, DS and DP release by the sponsor were reviewed:


**1. Sterility test of (b) (4) DP (Seth Schulte)**Introduction

Sterility testing is performed on (b) (4) Skysona™ DP at (b) (4). Acceptance criteria of 'No Growth' must be met for the lot release of Skysona™.

**Method**


(b) (4)

(b) (4)



Skysona™ DP Sterility Test Qualification

(b) (4)



Information Request and Review

An IR was sent to bluebird bio on 28 December, 2021 with responses received on 07 January, 2022 (Amendment #9) and 16 February, 2022 (Amendment #16). A second IR was sent to bluebird bio

on 15 April, 2022 and a response was received on 29 April, 2022 (Amendment #49). The IR response on 29 April, 2022 initiated an informal teleconference between bluebird bio and the FDA on 13 May, 2022 with a written follow up from bluebird bio on 27 May, 2022 (Amendment #61). A third IR was sent to bluebird bio on 29 June, 2022 and a response was received on 30 June, 2022 (Amendment #74). A fourth IR was sent to bluebird bio on 26 August, 2022 and a response was received on 31 August, 2022 (Amendment #106). These requests asked bluebird bio for clarification and additional data to supplement their sterility test reports VEN-TP-0177, VAL-VEN-RPT-0302, VAL-VEN-RPT-0377, and VAL-VEN-RPT-0514. The responses to CBER's IRs were reviewed as part of the DP sterility test qualification above.

Due to the unique nature of cellular and tissue products, there are challenges associated with accruing enough material for subsequent testing. DBSQC is aware strict adherence to testing standards for such products may not always apply due to limited production lot size. The main concern regarding smaller sample test volume for sterility testing is an increased potential for inadvertent infection associated with injection of contaminated product that may be detectable using larger sterility test sample volumes. (b) (4)

### Conclusion

The method suitability tests were performed and compliant with (b) (4) and the test results indicate there is no product inhibition on microorganism growth, thus indicating the (b) (4) sterility test method is appropriate under the actual conditions of use at (b) (4)

for Skysona™ DP

(b) (4)

## **2. Endotoxin test of (b) (4) DP (Seth Schulte)**

### Introduction

Endotoxin testing for

(b) (4)

. Acceptance criteria of

(b) (4)

for Skysona™ DP must be met for the lot release.

Method

The (b) (4) BET is a (b) (4) quantitative assay that (b) (4)

. The method is described in more detail below together with the tests that were performed to determine suitability of the test method.

(b) (4)

Skysona™ DP Bacterial Endotoxin Test Qualification

(b) (4)

Information Request and Review

An IR was sent to bluebird bio on 28 December, 2021 and a response was received on 07 January, 2022 (Amendment #9). This request asked bluebird bio for clarification and additional data to supplement their endotoxin test reports VAL-VEN-QUAL-0064 and VEN-RPT-0503. The response to CBER's IR was reviewed as part of the DP endotoxin test qualification above.

Conclusion

The method suitability tests were performed and compliant with (b) (4) and the test results indicate there is no product interference from the test sample, thus indicating the (b) (4) (b) (4) BET test method is appropriate under the actual conditions of use at (b) (4) facility for (b) (4) Skysona™ DP.

3. (b) (4) test of (b) (4) DP (Seth Schulte)  
(b) (4)

(b) (4)

(b) (4)








(b) (4)

(b) (4)

(b) (4)

3 pages have been determined to be not releasable: (b)(4)



(b) (4)



## **6. Appearance of (b) (4) DP (Tao Pan)**

### Introduction


The appearance of (b) (4) eli-cel DP were determined by visual inspection to assess their color, clarity, and particulate matter; the methods are based on (b) (4)



(b) (4)



(b) (4)



### *Appearance of DP*




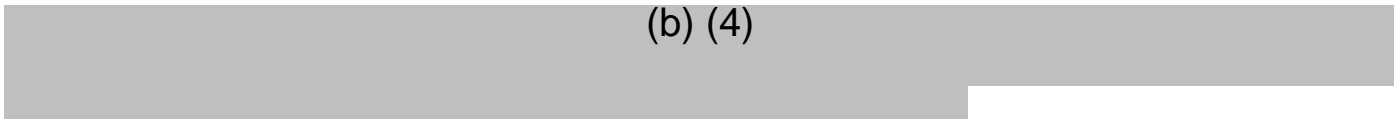
#### Method

The DP, eli-cel, is a cell suspension for infusion; for release, its appearance is defined as colorless to white to red, including shades of white or pink, light yellow, and orange. The appearance of eli-cel is assessed by visual inspection (b) (4) (SOP-Visual Appearance

(b) (4). In brief, DP sample in a (b) (4) was defined for the method. A detailed description of the method has been provided in the application.

#### Method Verification

(b) (4)



#### Conclusion

The appearance method for eli-cel DP is a simple method; based on information provided, it has been validated for its intended use of lot release testing of the eli-cel DP.

14 pages have been determined to be not releasable: (b)(4)