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1.0 Purpose

This SOP describes the preparation of End-of-Production (EOP) cell banks prepared from mammalian cell culture broth.

2.0 Scope

This procedure applies to personnel performing mammalian cell banks from stirred-tank bioreactors operations at the Biopharmaceutical Development Program (BDP)

3.0 Authority and Responsibility

3.1 The Director of Technical Operations, Biopharmaceutical Development Program, has the authority to define this procedure.

3.2 Production personnel are responsible for training personnel on this procedure and for documenting this training to Biopharmaceutical Quality Assurance (BQA).

3.3 Production personnel are responsible for the implementation of this procedure.

3.4 BQA is responsible for quality oversight of this procedure.

4.0 Guidelines

4.1 Refer to ***SOP 13200 - Qualification of Cells and CGMP Cell Banks****,* for general guidelines regarding cell banking and cell bank testing.

4.2 An EOP bank must be made for each GMP mammalian cell culture product.

4.3 A BDP part number must be established for the EOP bank being performed. This BDP part number will also be found on the associated Master Specification. If a BDP part number has not been created, one must be created.

4.4 The age of the cells used in preparing the bank must be the longest-running production during the campaign. If a subsequent production run is longer than the production from which the original EOP bank was prepared, another EOP bank must be prepared from this production.

4.5 The EOP bank processing should start within 2 hours of the time the sample was taken and may be stored at 2-8°C until it is vialed.

4.6 Testing of an EOP bank is generally not required for an early-phase product. If the EOP bank is required to be tested (by a regulatory agency, IND sponsor or NCI), a Master Specification will be generated and the EOP bank will be tested as per the Master Specification. General testing guidelines for EOP banks are given in ***SOP 13200 - Qualification of Cells and CGMP Cell Banks*.**

5.0 General Preparations

5.1 Clean and decontaminate the BSC to be used for the cell banking process per ***SOP 19102 - Routine Use and Disinfection of Biological Safety Cabinets, Incubators, Shakers, and Centrifuges***.

5.2 Clean the centrifuge using an approved disinfectant per ***SOP 19102 - Routine Use and Disinfection of Biological Safety Cabinets, Incubators, Shakers, and Centrifuges***, and set to the following parameters:

Temperature: 4°C

Speed: 100 RCF

Time: 10 minutes

6.0 Procedure

6.1 For all bioreactor productions: Working in the BSC, transfer ≥ 150 mL of the final production sample from which the EOP bank is to be prepared into a sterile 250mL PETG bottle. For shake flask productions: proceed to step 6.3, using the average viable count from the associated production run for the calculations.

6.2 Process the remainder of the final sample, per the (Master Production Record) MPR associated with this production run and use this final sample viable cell count for the following steps.

Perform the following calculations for EOP media preparation, depending on harvest source, and document on **Form 12210-01**, EOP Summary, (**Attachment 1**).

6.2.1 Volume of final production sample required for centrifugation:

6.2.1.1 Bioreactor Harvest

Desired total number of cells	/	Final sample viable cell count (cells/mL)	=	Volume production sample required (mL)
7.5×10^8	/		=	

6.2.1.2 Shake flask Harvest

Desired total number of cells	/	Average viable cell count (cells/mL)	=	Volume production sample required (mL)
7.5×10^8	/		=	

Volume production sample required (mL)	/	Number of shake flasks	=	Volume per flask required (mL)
	/		=	

6.2.2 Volume of CryoStor, (PN: 10617), required is 75mL.

6.3 Aseptically pipette the volume of sample required for centrifugation equally between sterile 50 mL centrifuge tubes.

6.4 Process the sample in the centrifuge setup in step 5.2.

6.5 Aseptically decant or pipette the supernatant (conditioned medium) from the centrifuged tubes.

6.6 Obtain a sterile 250 mL PETG bottle or equivalent for the EOP bank container.

6.7 Pipette the required volume of CryoStor, pn: 10617, into the cell pellet containers, resuspend via pipetting and add this to the EOP bank container.

6.8 Mix the EOP bank by swirling the bottle or pipetting until thoroughly mixed for 1 – 5 minutes and record on attachment.

6.9 Fill cryovials with 1 ± 0.2 mL of the EOP bank as follows.

6.9.1 Using aseptic technique, remove the cap from each cryovial and, using a pipette, transfer 0.8-1.2 mL of the cell suspension into the cryovial, then re-cap the cryovial and place the vial in the storage box. Fill a minimum of 50 vials. Swirl the EOP bank periodically to keep the cell suspension mixed throughout vialing process.

6.9.2 Label the cryovials "EOP bank," product, lot number, and part number, date and initials.

6.9.3 Store the prepared cell bank vials at 2-8°C for no more than 4 hours until they are transferred for slow rate freeze, ***SOP 12208 – Operation of the Thermo 7450 Controlled Rate Freezer***. Attach record of slow rate freeze as part of **Attachment 1, Form 12210-01**.

6.10 Deliver vials after the freeze to MMIC for storage at ≤ -70°C per ***SOP 20303 - CGMP Product Accountability***.

6.11 The project scientist will withdrawal necessary samples from MMIC and submit to PA/QC for testing per the Master Specification and ***SOP 22002 - Request for Quality Control Testing***.

7.0 Documentation

7.1 Document this procedure on **Form 12210-01**, EOP Banking Summary.

7.2 Attached the completed **Form 12210-01** to the batch record pertaining to the lot from which the EOP bank was prepared.

8.0 References and Related Documents

SOP 13200	*Qualification of Cells and CGMP Cell Banks*
SOP 19102	*Routine Use and Disinfection of Biological Safety Cabinets, Incubators, Shakers, and Centrifuges*
SOP 12208	*Operation of the Thermo 7450 Controlled Rate Freezer*
SOP 20303	*CGMP Product Accountability*
SOP 22002	*Request for Quality Control Testing*
Form 12210-01	*EOP Bank Summary*