STANDARD OPERATING PROCEDURE	
Department: Microbiology	**SOP No.:**
Title: Receipt, Incubation, Observation & Discard of Media Fill Vials & Inoculated Membrane	**Effective Date:**
Supersedes: Nil	**Review Date:**
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1.0 OBJECTIVE

1.1 To lay down the procedure for receipt, incubation, observation & discard of media fill vials & inoculated membrane.

2.0 SCOPE

2.1 This procedure is applicable for Microbiology Laboratory.

3.0 RESPONSIBILITY

3.1 Microbiologist is responsible for receipt, incubation, observation & discard of media fill vials & inoculated membrane.

4.0 ACCOUNTABILITY

4.1 Head Microbiology

5.0 EHS CONSIDERATIONS

5.1 NA

6.0 PROCEDURE

6.1 **Receipt and incubation of media fill vials & inoculated membrane:**

6.1.1 Media fill vials & Inoculated membrane flask shall be transferred through dynamic pass box of production area to microbiology laboratory.

6.1.2 Media fill vial should be incubated in the validated BOD incubators.

6.1.3 Prior to incubation the vials with the microbiological growth medium should be inverted or otherwise manipulated to ensure that all surfaces, included the internal surface of the closure, are thoroughly wetted by the medium.

6.1.4 Count the total number of vials of and record the details in Annexure II.

6.1.5 It is generally accepted to incubate at 22.5±2.5°C for a minimum of 7 days followed immediately, or after a first reading, by incubation at 32.5±2.5°C for a total minimum incubation time of 14 days

6.1.6 Incubate all vials of media fill 22.5±2.5°C for initial 7 days.

6.1.7 After completion of 7 days of incubation, transfer the vials at 32.5±2.5°C for next 7 days.

6.1.8 Incubate the vials in upright in initial 07 days at 22.5±2.5°C and in inverted position next 07 days at 32.5±2.5°C.

6.1.9 In this way, vials will be incubated upright and inverted position at 22.5±2.5°C and 32.5±2.5°C.

6.1.10 Inoculated membrane flask shall be incubated for 14 days as FTM flask at 32.5±2.5°C & SCDM flask at 22.5±2.5°C.

6.2 **Observation of media fill vials & inoculated membrane:**

6.2.1 The media fill vials shall be inspected at minimum interval of 7 & 14 days of incubation and result shall be recorded in batch record of Media fill as well as in Annexure II.

6.2.2 Each person performing inspection shall not perform inspection for more than 2 hours at a stretch shall take at least 15 minutes rest after every 2 hour of inspection.

6.2.3 Personnel should be trained for this task.

6.2.4 Each media fill vial should be examined by trained microbiologist on completion of 7 days incubation of both the temperature conditions incubation for turbidity and if turbidity is observed in any vial it should be reported to QA immediately.

6.2.5 When inspecting the vials they should be compared to a known sterile vial for comparison as some microbial growth shows up as a faint haze which is difficult to detect unless there is a control vials to compare against.

6.2.6 Pick a single vial without obstructing view field by the hands holding from top.

6.2.7 Invert the vials and gently swirl them and look for microbial growth.

6.2.8 If any vial with microbial growth/turbidity is found keep the vial in rejection tray with proper identification and record the details in Annexure II.

6.2.9 The microorganisms present in the vial of the simulation test should be identified to genus but preferably species level to aid determination of the possible sources of the contamination.

6.2.10 Inoculated membrane flask shall be observed daily and details shall be recorded in Annexure I.

6.2.11 Perform post GPT & container closure integrity after completion of 14 days of incubation of media fill vials.

6.3 **Discard of media fill vials & inoculated membrane:**

6.3.1 After completion of fourteenth day inspection media fill vials shall be removed from BOD incubator.

6.3.2 Collect the remaining vials in poly bag and label the bag as 'Media fill vials for Incineration-Biohazard'.

6.3.3 All the poly bags shall be sent to the external party for incineration.

6.4 **Discard of contaminate media fill vials & inoculated membrane:**

6.4.1 After identification of contaminated media fill vials & inoculated membrane, discard the media fill vials & inoculated membrane through as per SOP of disposal of microbial waste SOP.

7.0 DEFINITIONS AND ABBREVIATIONS

7.1 GPT - Growth Promotion Test

7.2 BOD - Biological Oxygen Demand

7.3 SCDM - Soybean casein digest medium

7.4 FTM - Fluid thioglycollate medium

8.0 REFERENCE

8.1 PICS, Recommendation on the validation of aseptic processes

9.0 ANNEXURES

9.1 Annexure I : Report for Inoculated membrane

9.2 Annexure II : Receipt, incubation and observation of media fill vials

10.0 DISTRIBUTION DETAILS

10.1 Controlled copy of this SOP shall be distributed to Quality Assurance, Production and Microbiology.

11.0 REVISION HISTORY

Supersedes SOP No.	Change Control No.	Reason for revision

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ANNEXURE I
REPORT FOR INOCULATED MEMBRANE

Name of Product				
Batch No.				
Inoculated by /Date				
Incubated by /Date				
	Date	**Time**	**BOD Incubator ID**	**Incubation temperature**
SCDM flask containing filter membrane				22.5±2.5°C
FTM flask containing filter membrane				32.5±2.5°C

Media Details				
Name of Media	**Sterile Lot No.**	**pH**	**Volume**	**Result of GPT**
SCDM				Pass / Fail
FTM				Pass / Fail

Observations														
Medium	**Days**													
	1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**14**
SCDM														
FTM														
Negative Control														
Observed by / Date →														
Checked by / Date →														

Expression of observation: 'NG' for 'No Growth', 'G' for 'Growth'

Remarks: The aseptic process simulation complies / does not comply as per specifications.

Checked by:
Date:

Reviewed By:
Date:

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ANNEXURE II
RECEIPT, INCUBATION AND OBSERVATION OF MEDIA FILL VIALS

Media fill for				
Media fill batch No				
Total number of media fill vials received				
Received on date & time				
Received by				
	Date	**Time**	**BOD Incubator ID**	**Incubation temperature**
Vials incubation started on				22.5±2.5°C
Vials incubation completed on				22.5±2.5°C
Vials incubation started on				32.5±2.5°C
Vials incubation completed on				32.5±2.5°C

Incubation day	**Date**	**Time**		**Number of contaminated vials**	**Observed by**
		From	**To**		

Remarks: The media fill complies / does not comply as per specifications.

Checked by:
Date:

Reviewed By:
Date: