PHARMA DEVILS

MICROBIOLOGY DEPARTMENT

STANDARD OPERATING PROCEDURE

Title: Microbiological Environmental Monitoring				
SOP No.:		**Department:**	Microbiology	
		Effective Date:		
Revision No.:	00	**Revision Date:**		
Supersede Revision No.:	Nil	**Page No.:**	1 of 12	

1. **Purpose:** The Purpose of this SOP (Standard Operating Procedure) is to describe the procedure for Microbiological Environmental Monitoring in the Factory premises.

2. **Scope:** This SOP is applicable to environmental monitoring carried out at different sections in non sterile drug product manufacturing areas like Sampling area,Tablet manufacturing area, Capsule manufacturing. Area, QC Microbiology area.

3. **References, Attachments & Annexures:**
   - 3.1 **References:**
     - 3.1.1 USP
     - 3.1.2 IP
     - 3.1.3 BP
     - 3.1.4 Schedule M
     - 3.1.5 SOP:Receipt,Storage,Preparation,Growth Promotion Test,Use And Disposal of Microbiological Media
   - 3.2 **Attachments:**
     - 3.2.1 Attachment-1: Environmental Monitoring analytical worksheet
     - 3.2.2 Attachment-2: Action plan for out of alert limit results of environmental monitoring
     - 3.2.3 Attachment-3:Action plan for out of action limit results of environmental monitoring
   - 3.3 **Annexure:**
     - 3.3.1 Annexure-1:Environmental Monitoring Schedule

4. **Responsibilities:**
   - 4.1 **Microbiologist:**
     - 4.1.1 To perform the activity as per SOP.
     - 4.1.2 To maintain the records as per SOP.
   - 4.2 **QC Head or designee:**
     - 4.2.1 To check the SOP.
     - 4.2.2 To give training to all concern persons..
   - 4.3 **Quality Assurance:**

4.3.1 To check the SOP

4.3.2 To ensure proper implementation of SOP.

4.4 **Regulatory Affairs, Quality Head , Plant Head:**

4.4.1 To review and approve the SOP.

5. Distribution:

5.1 Quality Assurance

5.2 Quality Control

5.3 Production department

5.4 Warehouse

5.5 Maintenance Department.

6. Abbreviations & Definition of Terms:

6.1 **Abbreviations:**

6.1.1 **EM** : Environmental Monitoring

6.1.2 **SCDA** : Soyabean Casein Digest Agar

6.1.3 **CFU** : Colony Forming Unit

6.1.4 **GPT** : Growth Promotion Test

6.1.5 **SPC** : Settling Plate Count

6.1.6 **LT** : Less than

6.1.7 **NMT** : Not More Than

6.1.8 **OOS** : Out of specification

6.1.9 **LAF** : Laminar Air Flow

6.1.10 **S.S.** : Stainless Steel

6.1.11 **F.B.D** : Fluidized bed dryer

6.1.12 **RODAC** : Replicate organism detection and counting

6.2 **Definition of Terms :**

6.2.1 **Alert Level /limit:** Levels or ranges which,when deviated from,signal a potential drift from operating conditions, these ranges are not perceived as being detrimental to end product quality.

6.3 **Action level /limit:** Levels or ranges distinct from product specification, which, when deviated from,signal a drift from normal operating conditions and require action.

7. Procedure:

7.1 **Media for Environmental Monitoring:** Pre-incubated plates of Soyabean Casein Digest agar media shall be used.

7.2 **Media preparation,GPT and Pre-incubation of media plates:** Perform the activity as per SOP "Receipt,Storage,Preparation, Growth Promotion Test, Use and Disposal of Microbiological Media" shall be followed.

7.3 **Frequency for EM:** Environmental monitoring shall be performed at least once in a month for non sterile manufacturing areas, primary packaging areas, dispensing areas, sampling areas , sex hormone manufacturing area and in microbiology laboratory plates shall be exposed weekly.

Note : Environmental monitoring frequency can be increased as per requirement of individual location.

7.4 **Environmental Monitoring schedule , sampling area and sampling locations :**

7.4.1 Microbiologist shall prepare the environmental monitoring schedule as per defined location.

7.4.2 Area where environmental monitoring needs to be done shall be defined in SOP and accordingly environmental monitoring schedule shall be prepared.

7.4.3 Sampling locations for settle plates in clean room should include areas where there is little air movement (i.e Dead spaces) or where air flow converge (come together or towards the same point or approach from different directions). Areas where these conditions are most likely to occur are,

7.4.3.1 Adjacent to doors.

7.4.3.2 At low level return air grilles.

7.4.3.3 In corners of rooms.

7.4.3.4 Any other place depending upon criticality of operation.

7.4.4 The plates shall be exposed 6 inches away from the return riser and in clockwise direction in each cubicle..

7.4.5 In each cubicle four plates shall be exposed in each of the four corners and it's location numbering system shall be as followed:

7.4.5.1 G01, G02------- Gnn for all areas of Ground floor.

7.4.5.2 F01, F02------- Fnn for all areas of First floor.

7.4.5.3 S01,S02-------Snn for all areas of Second floor.

7.4.5.4 UG01,UG02 -----UGnn for all areas of Upper ground floor.

7.4.5.5 UB01,UB02 ------UBnn for all areas of Upper basement floor (Sex hormone area)

7.4.5.6 GMP01,GMP02--GMPnn for corridor of Ground floor main passage

7.4.5.7 FMP01, FMP02---- FMPnn for corridor of First floor main passage

7.4.5.8 FLS01, FLS02---- FLSnn for corridor of First floor left side

7.4.5.9 FRS01, FRS02---- FRSnn for corridor of First floor right side

7.4.5.10 SMP01,SMP02----SMPnn for corridor of Second floor.main passage

7.4.5.11 SLS01,SLS02-------SLSnn for corridor of Second floor left side

7.4.5.12 SRS01,SRS02-------SRSnn for corridor of Second floor right side

7.4.5.13 UGMP01,UGMP02 -----UGMPnn for areas of Upper ground floor main passage

7.4.5.14 UBMP01,UBMP02 ------UBMPnn for areas of Upper basement floor main passage (Sex hormone area)

Where nn is serial number of area.....

7.5 **Environment monitoring by settle plate method:**

7.5.1 Examine the pre-incubated plates for any contamination.

7.5.2 Mark (label) the plates with location number as per schedule , media name, analyst sign, date and time of exposure.

7.5.3 Before taking petri plates into area, microbiologist shall fill the details like Media preparation lot no, area name, exposure time in environmental monitoring analytical worksheet as per attachment 1.

7.5.4 Microbiologist shall carry the pre-incubated plates in the S.S. container which is previously sanitized with 70% IPA solution.

7.5.5 In non sterile area plates shall be exposed preferably in operation condition for 4 hours . If count for 4 hours is uncountable as per trend data the exposure time can be decreased.

7.5.6 At the time of exposure take out the petri plates from SS container for exposure.

7.5.7 Place the petri plates with its lid at predetermined points of environmental monitoring as per the schedule.

7.5.8 Remove the lid of petri plate and rest it on the dish base and expose the plate for minimum 4 hours.

7.5.9 After exposure, carefully close the petri plates with its lid from all location and keep the plates back into the SS container and transfer the plates to microbiology lab for incubation.

7.6 **Incubation of plates:**

7.6.1 Microbiologist shall ensure that all petri plates are incubated within 4 hours from the time plate exposure completed.

7.6.2 Incubate the all exposed plates at 20–25 °C for 72 hours along with positive and negative control plate in inverted position.

7.6.3 Further incubate the same plates in an inverted position at 30-35°C for another 48 hrs. along with positive and negative control plate.

Note : For positive control, record the observation from GPT record and use pre-incubated plate as negative control. Positive control must show growth. Negative control must not show any growth. If no growth in positive control or growth observed in negative control refer system of investigation

7.7 **Observation:**

7.7.1 After the incubation observe the plates for bacterial and fungal count and note down the observations in environmental monitoring analytical worksheet as per attachment-1.

7.7.2 The total no. of colony forming unit (CFU) shall be calculated as a sum of bacterial (B) CFU and fungal (F) CFU.

7.8 **Acceptance criteria: For settle plate (In operation limits)**

7.8.1 Limit: NMT 100 cfu/plate .

7.8.2 Alert limit : NMT 70 cfu/plate.

7.8.3 Action limit: NMT 90 cfu/plate.

7.8.4 For Class 100 Area: Less than 1 CFU/plate,

7.8.5 For non classified area alert and action limits shall be established based on sufficient trend data. Also limit shall be reviewed on yearly basis and revised if required.

Note: For non classified area, whenever the activity is started first time, the limit in SOP can be kept as "To be established based on trend data".

7.9 **Acceptance criteria: For RODAC plate**

7.9.1 Limit: NMT 50 cfu/plate

7.10 **Handling of out of limit results:**

7.10.1 Periodically conduct a document review of the environmental monitoring results and trends. The documentation of the review must include the action to be taken,if any.

7.10.2 Microbiologist shall intimate to QC Head ,QA department and production department for any type of crossing of alert and action limit.

7.11 **For Alert Limit:**

7.11.1 Microbiologist shall send intimation to production department through QA in case of out of alert limit results as per attachment-2.

7.11.2 Production supervisor shall carry out investigation as per following check points (but not limited to those) and return the form to microbiologist for recommendation.

7.11.3 Microbiologist shall review the investigation details and shall recommend the corrective action.

7.11.4 Production shall take corrective action as per recommendations and shall return the form to microbiologist.

7.11.5 After corrective action taken, microbiologist shall carry out the environment monitoring of that location for three consecutive days and if results found above the alert limit then action shall be taken based on the finding of investigation.

7.12 **For Action Limit:**

7.12.1 Microbiologist shall send intimation to production department through QA in case of out of alert limit results as per attachment-3.

7.12.2 The action level also appears in case of any particular species is established in to the area e.g. Fungus, gram negative bacilli etc..

7.12.3 Detail investigation shall be carried out by the Production supervisor, QA and Microbiologist, which shall be reviewed by Quality Head.

7.12.4 Based on finding of the investigation, decision to continue or discontinue the production activity shall be taken.

7.12.5 Impact on product / activities carried out during the span of investigation shall be studied. as a part of that, microbial limit test of finish product shall be done in case if the results of environment monitoring are found out of action limit.

7.12.6 After corrective action Microbiologist shall carry out the environment monitoring of that position for three consecutive days and if results found above the action limit then operation will be stopped and investigation shall be carried out. Action shall be taken based on the finding of investigation.

7.12.7 **Check list for investigation of out of limit results:**

7.12.7.1 Area cleaning record.

7.12.7.2 Area sanitization record.

7.12.7.3 Processing records (e.g. For any incidences like spoilage of water bursting of FBD bag, prolonged power failure.)

7.12.7.4 Equipment / instrument usage log (maintainable work carried out in area).

7.12.7.5 Differential pressure of the area /room.

7.12.7.6 Temperature / Humidity of the area or room.

7.12.7.7 Any deviation.

7.12.7.8 Trends of Environmental monitoring results for the production area.

7.12.7.9 Review training records for all individuals in the activity to ensure that proper training was provided.

Attachment – 1
Environmental Monitoring analytical worksheet

Area Name		Exposure Time : From_____To_____	**Date of Exposure**	_____

Media	Soyabean Casein Digest Agar	Media Preparation Lot No:_____	**Pre-incubation Date**	_____
Incubation	Incubation at 20 to 25 °C for 72 Hrs Temperature :_____ Incubator No:_____ Incubation in Date : _____ Incubation out Date :_____		Incubation at 30 to 35 °C for 48 Hrs Temperature :_____ Incubator No:_____ Incubation in Date : _____ Incubation out Date :_____	

SETLLING PLATE COUNT

Location No.	Location Name and Activity	B (cfu/plate)	F (cfu/plate)	Total count/Plate (B+F)

***Acceptance Criteria :** Limit: 100 cfu/plate

For Class 100 Area: Less than 1 CFU/plate (**Abbreviations:** B = Bacterial count and F = Fungal count)

Positive Control:	Negative control:
Satisfactory /Unsatisfactory	Satisfactory /Unsatisfactory

Exposed By	**Observed By**	**Checked By**	**Approved By**
Date:	**Date:**	**Date:**	**Date:**
Date of exposure		Date of intimation	
Area		Intimated By	

S.No.	Location	Findings	Limit

Investigation Details:

Check Points	Reviewed by concern Department Head	Checked by (Microbiologist)	Verify by (QA)

Area cleaning record			
Area sanitization record.			
Processing records (e.g. For any incidences like spillage of water,bursting of FBD bag, prolonged power failure etc.)			
Equipment / instrument usage log (maintainence work carried out in area).			
Differential pressure of the area /room.			
Temperature / Humidity of the area or room.			
Any deviation.			
Trends of environment monitoring.			
Review of training records.			

Recommendation:

Given By / Date
(Microbiologist)

Approved By/Date
(QC Head)

Corrective action taken:

Taken By/Date
(Production supervisor)

Verified By /Date
(Production Head)

Supportive data for corrective action:

Attachment - 1:

Attachment – 2:

Checked By / Date:_____ **Approved By/ Date:_____**

(Microbiologist) **(QC Head)**

Attachment –3

Action plan for out of action limit results of environmental monitoring

Date of exposure		**Date of intimation**	
Area		**Intimated By**	

S.No.	Location	Findings	Limit

Investigation Details:

Check Points	Reviewed by concern department head	Checked by Microbiologist	Verify by QA
Area cleaning record			
Area sanitization record.			
Processing records (e.g. For any incidences like spillage of water , bursting of FBD bag, prolonged power failure.)			
Equipment / instrument usage log (maintainable work carried out in area).			
Differential pressure of the area /room.			
Temperature / Humidity of the area or room.			
Any deviation			
Trends of environment monitoring.			
Review of training records.			

Recommendation:

Given By /Date
(Microbiologist)

Approved By/Date
(QC Head)

Corrective action taken:

Taken By/Date
(Production supervisor)

Verified By/Date
(Production head)

Supportive data for corrective action:

Attachment - 1:

Attachment – 2:

Checked By/Date:_____

(Microbiologist)

Approved By/Date:_____

(QC Head)

Annexure-1/ 00
Environmental Monitoring Schedule

Location Name	Location No.	Plate Exposure Date	Frequency

Prepared By:	Checked By:	Approved By:

8. History:

Version No.		Effective Date	