PROTOCOL
FOR
CLEANING VALIDATION
PRODUCTION LYOPHILIZATION

TABLE OF CONTENTS

1. OVERVIEW:

1.1 Introduction:

The purpose of this protocol for assurance of cleaning methodology to detect previous product residue from equipment/equipments parts. To provide the necessary documentary evidence that the cleaning procedure can reduce the residual contamination of previous product below the established limit so that such residue do not affect the quality and safety of the subsequent product to be manufactured in the same equipment.

Cross contamination risk:-

Production Lyophilization is a product dedicated facility established in a carbapenem dedicated premises for producing Ertapenem for Injection 1g/vial (Lyophilized) of carbapenem family. Hence the risk of cross contamination with other product is negligible.

In this facility the only risk is, if inefficient cleaning of the product residue which may remain on to the used equipment surface and this residue can be further degraded in to the unknown organic matter and a source of impurity carries forward to the next batch of the product during manufacturing.

1.2 Objective:

The objective of the cleaning validation protocol is to establish & assure with documented evidence that the cleaning procedure for cleaning of process equipments (Mixing vessel, filtration line, Holding vessel, Buffer tank and Filling Nozzle/piston including Equipment connection lines) are reproducibly remove residue of the previous product to levels below the established acceptance limits which may be soiled and further degraded and cause of Physical, Chemical, Biological contamination in next product batches.

Though this is dedicated facility for Ertapenem molecule but batch to batch cleaning subjected for validation.

- To verify that the cleaning procedure is effectively capable for removal of residual product/degrading product.
- To eliminated any risk of product degrading in to subsequent batches.

1.3 Scope:

The scope of this validation activity is limited to validate the cleaning process equipment (Mixing vessel, filtration line, Holding vessel, Buffer tank and Filling Nozzle/piston including Equipment connection lines) in production lyophilization department.

1.4 Responsibility:

To conduct the cleaning validation study, a team shall be formed. The validation team shall contain the members from the Quality Control, Engineering, Production and Quality Assurance Departments.

The Validation team is described through the following responsibility:

Quality Assurance	To prepare the protocol & report
	To provide the training
	To execute & supervise the study

Quality Control, Engineering, Production and Quality Assurance	To review the protocol
	To conduct the study
	To collect and analyze samples

Reviewer-1 : QC In-Charge / Designee	To review validation protocol. To review validation report.
Reviewer-2: Validation - CQA (SME)	To review validation protocol. To review validation report.
Approver-1: Head Production /Designee	To approve validation protocol. To approve validation report.
Approver-2: Head-QA/Designee	To approve validation protocol. To approve validation report.

2. EXECUTION TEAM:

Following personnel shall be responsible for the execution of validation study:

Production	:	To conduct the validation study as per protocol.
Quality Assurance	:	To collect the sample as per protocol.
Quality Control	:	To conduct the microbiology monitoring, collect & analysis of samples and reporting of results.

3. TRAINING RECORD:

3.1 **Purpose:**

The purpose of the training is to familiarize the trainees with the purpose and procedure of cleaning validation activity.

3.2 **Scope:**

This training is applicable to the protocol for cleaning validation.

3.3 **Topics:**

The following topics shall be covered during training: Identifying the responsibility of involved person.

4.3.1 Purpose & procedure of cleaning validation.

4.3.2 Documentation practices to be followed.

4.3.3 General precautions / guidelines to be followed during validation.

4. REQUIREMENT FOR CLEANING VALIDATION:

4.1 Documental Requirements:

S.No.	Document No.	Title of SOP
1.		Procedure for cleaning, arranging & wrapping of filling machine parts
2.		Cleaning of stainless steel vessels lines and filtration train
3.		Procedure for cleaning of Lyophilizer.
4.		Procedure for calibration of high performance liquid chromatography.
5.		Procedure for operation of high performance liquid chromatography (Model-Shimadzu).
6.		Procedure for operation of high performance liquid chromatography (Model-Waters).
7.		Bacterial endotoxin test procedure.
8.		Particulate matter testing.
9.		Bio burden testing by filtration method.
10.		Water testing procedure.

4.2 Apparatus / Instrument Requirements:

Test tube (Glass bottle), WFI, Rinse Sample bottle, Depyrogenated bottles, Autoclaved bottles.

5. SYSTEM/EQUIPMENT DESCRIPTION:

5.1 API Solubility:

5.1.1 **Ertapenem**: Freely soluble in water

5.2 Solubility Matrix:

DESCRIPTION	SOLUBILITY (1g in listed ml)	mg/ml
Very soluble (VS)	Less than 1	>1000
Freely soluble (FS)	From 1 to 10	100 to 1000
Soluble (S)	From 10 to 30	33.33 to 100
Sparingly soluble (SPS)	From 30 to 100	10 to 33.33
Slightly soluble (SLS)	From 100 to 1000	1 to 10
Very slightly soluble (VSLS)	From 1000 to 10 000	0.1 to 1
Practically insoluble (PI)	More than 10 000	<0.1

5.3 **Equipment Description:**

Following equipments are directly contacts with product-

S.No.	Equipment Name	Sampling method	Justification
1.	Mixing vessel	Rinse	Mixing vessel is closed with lid and it is not opened during cleaning, hence swab sampling is not possible and rinse sampling to be taken.
2.	Holding vessel	Rinse	Holding vessel is closed with lid and it is not opened during cleaning, hence swab sampling is not possible and rinse sampling to be taken.
3	Transfer lines	Rinse	Transfer lines surface area is not appropriate for swab sampling, hence rinse sampling to be taken
4	Buffer tank	Rinse	Buffer tank is closed with lid and it is not opened during cleaning, hence swab sampling is not possible and rinse sampling to be taken.
5	Machine parts i.e. Manifold, piston and shaft	Rinse	Machine parts are irregular shape and not suitable for swab sampling, hence rinse sampling to be taken.

6. CLEANING VALIDATION METHODOLOGY OR PROCEDURE:

6.1 **General Recording Instructions:**

6.1.1 Read the contents of the document thoroughly before proceeding for Execution of the activity (in case of doubts / contradictions / contact the approvers of the document for clarifications).

6.1.2 Recording of all the observations and data shall be done as per **SOP "Good Documentation Practices"**.

6.2 **Cleaning Methodology:**

6.2.1 After completion of equipment cleaning Production and QA personnel shall visually inspect equipment internal/external surfaces to verify the cleanliness and record the observation.

6.2.2 After visual inspection, if found satisfactory rinse the equipment as per respective SOP.

6.3 **Worst case product selection:**

6.3.1 Worst case product shall be selected by following criteria-

6.3.1.1 10 ppm criteria

6.3.2 **Product matrix:**

Table-1

S.No.	Name of product	API	Therapeutic Dose Minimum (TD) (mg)	Therapeutic Daily Dose Maximum (TDD)(mg)	Minimum Batch Size	Solubility (in water)
1.	Ertapenem for Injection 1g / vial	Ertapenem	1000	1000	25.00 (Ltrs.)	Freely soluble

6.3.3 **Maximum Allowable Residue (MAR) calculation:**

6.3.3.1 **10 ppm Criteria:** General limit for maximum allowed concentration (mg/mg or ppm) of "previous" Substance in the next batch. = **0.00001mg** or 10 ppm.

☞ *Cleaning validation study the MACO (Maximum allowable carry over) limit based on 10ppm criteria and Dose criteria is not considered due to single product manufacturing facility.*

6.4 **Sampling Procedure and Test Methodology:**

6.4.1 **Physical verification:**

6.4.1.1 After completion of cleaning Production and QA personnel shall physically verify the equipment for cleanliness. It should be visually clean then only further activity shall be performed.

6.4.1.2 During rinsing collect the rinse sample as per sampling procedure from the sampling location defined in **Table -1** and send the sample to QC department for analysis.

6.4.1.3 Also collect the water use for rinsing in a separate container and send the sample to QC department for analysis as a blank.

6.4.1.4 Properly label the each sample container with the information of Equipment ID/Sample location, Sample name, Date /time of sample and sampled by.

6.4.2 **Rinse Sample:**

<table>
<tr><th colspan="8">Table-1</th></tr>
<tr><th rowspan="2">S. No.</th><th rowspan="2">Name of Equipment</th><th rowspan="2">Equipment ID</th><th rowspan="2">Contamination risk consideration</th><th rowspan="2">Cleaning SOP No.</th><th colspan="2">Sampling consideration</th><th rowspan="2">Testing Consideration</th></tr>
<tr><th>Sample Method rational</th><th>Sample location/Rinse Volume</th></tr>
<tr><td>1.</td><td>Scott bottle</td><td>NA</td><td rowspan="4">Not product contact, used only for preparation, cooling and holding the NaOH solution in WFI for pH adjustment of Product during process Hence consider non critical for cleaning validation study</td><td rowspan="4">AN-PR-PL-CLN-0011</td><td>NA</td><td>Final Rinse Sample from Mouth/100 ml</td><td rowspan="5">• BET
• Particulate Matter</td></tr>
<tr><td>2.</td><td>Measuring cylinder</td><td>NA</td><td>NA</td><td>Final Rinse Sample from Mouth/100 ml</td></tr>
<tr><td>3.</td><td>NaOH vessel</td><td></td><td>NA</td><td>Final Rinse Sample from drain/5Litre</td></tr>
<tr><td>4.</td><td>Cooling vessel</td><td></td><td>NA</td><td>Final Rinse Sample from drain/20Litre</td></tr>
<tr><td>5.</td><td>Transfer line-Cooling vessel to Mixing vessel</td><td>NA</td><td>NA</td><td>NA</td><td>NA</td><td>Final Rinse Sample from drain the of mixing vessel/ 20Litre</td></tr>
<tr><td>6.</td><td>Mixing vessel</td><td></td><td>In Product contact used for compounding of drug, Hence consider critical for cleaning validation study</td><td></td><td rowspan="2">Cleaning and Rinsing of entire exposed internal vessel surface by WFI through spray ball Flushed/rinse the entire internal surface of vessel to get rod the stick material residue from equipment surface and the rinse sample from end point can be considered as a representative sample for estimation of residual leftover.</td><td>Final Rinse Sample from drain/20Litre</td><td>• Residue of previous product
• BET
• Particulate Matter</td></tr>
<tr><td>7.</td><td>Holding vessel</td><td></td><td>In Product contact used for Holding the product solution (dissolved drug in WFI),Hence consider critical for cleaning</td><td></td><td>Final Rinse Sample from drain **20**Litre</td><td>• Residue of previous product
• BET
• Particulate Matter</td></tr>
</table>

Table-1

S. No.	Name of Equipment	Equipment ID	Contamination risk consideration	Cleaning SOP No.	Sampling consideration		Testing Consideration
					Sample Method rational	Sample location/Rinse Volume	
			validation study				
8.	Transfer line from mixing vessel to holding vessel through Filtration train-1	NA	In product contact used for transfer of product solution from Mixing vessel to holding vessel hence consider critical for cleaning validation study		Entire internal surface area of transfer line exposed to WFI during flushing/rinsing of line to clean and the rinse sample from end point can be considered as a representative sample for estimation of residual leftover.	Final Rinse Sample from drain the of Holding vessel/ **20**Litre	• Residue of previous product • BET • Particulate Matter
9.	Transfer line from holding vessel to Buffer tank and Filling machine transfer line through Filtration train-2	NA				Final Rinse Sample from drain the of buffer tank/ **20**Litre.	• Residue of previous product • BET • Particulate Matter
10.	Buffer Tank	NA	In product contact used for filling the product		Entire internal surface area of transfer line exposed to WFI during flushing/rinsing of line to clean and the rinse sample from end point can be considered as a representative sample for estimation of residual leftover.	Final Rinse Sample from drain the of buffer tank/ 5Litre.	• Residue of previous product • BET • Particulate Matter
11.	Filter cartridge	NA	Every Time new filter used hence not consider in this cleaning validation study.				
	Machine		In product contact used for filling		Entire internal/external surface	Final Rinse Sample/2 Liter	• Residue of previous product

Table-1

S. No.	Name of Equipment	Equipment ID	Contamination risk consideration	Cleaning SOP No.	Sampling consideration		Testing Consideration
					Sample Method rational	Sample location/Rinse Volume	
12.	Parts – Manifold	NA	the product solution, Hence consider critical for cleaning validation study		area of manifold exposed to WFI during flushing/rinsing to clean the line internal/external surface hence no hard to clean area considered		• BET • Particulate Matter
	Piston, shaft	NA	In product contact used for filling the product solution, Hence consider critical for cleaning validation study		Entire internal/external surface area of nozzle/piston exposed to WFI during flushing/rinsing to clean the line internal/external surface hence no hard to clean area considered	Final Rinse Sample/2 Liter	• Residue of previous product • BET • Particulate Matter
13.	Silicon tubes	NA	Every Time new clean tube used hence not consider in this cleaning validation study.				
13.	Rubber stopper bowl, chute, rotary plate	NA	Not product contact, only used for rubber stoppering system Hence consider non critical for cleaning validation study		Entire internal surface area of transfer line exposed to WFI during flushing/rinsing	Final rinse sample from internal surface of the Rubber stopper bowl. /100 ml	• BET • Particulate Matter
14.	NaOH addition Funnel	NA	Not product contact surface		NA	NA	NA
15.	Lyophilizer		Not product contact used for holding the filled half rubber stopper vials for Lyophilization		Entire internal surface area of Lyophilizer and its line is cleaned through automated CIP system	Final Rinse Sample from drain the of Lyophilizer collected during Cleaning/	• BET • Particulate Matter

Table-1

S. No.	Name of Equipment	Equipment ID	Contamination risk consideration	Cleaning SOP No.	Sampling consideration		Testing Consideration
					Sample Method rational	Sample location/Rinse Volume	
			on the shelves validation study.			100 ml	

6.5 **Critical Step of equipment cleaning procedure:**

6.5.1 **Sodium Hydroxide Vessel:**

6.5.1.1 Sodium Hydroxide preparation vessel inner surface is flushed with jet valve WFI for 1 min flushed WFI drained through the bottom of the vessel.

6.5.1.2 Aging flushed the vessel with WFI and allows filling the WFI up to the neck level.

6.5.1.3 Drain the whole WFI to the drain.

6.5.1.4 Again flushed the vessel with 10 Ltrs. of WFI and with draw the rinse sample for analysis.

6.5.2 **Cleaning of Sodium hydroxide addition funnel:**

6.5.2.1 Flush the inner surface wall with WFI by rotating slowly.

6.5.2.2 Filled the WFI in the addition funnel up to the over flow level and drain it twice.

☞*Ensure physically there are no spots on the inner surface.*

6.5.3 **Dry powder charging funnel:**

6.5.3.1 Flush the inner surface wall with WFI by rotating slowly twice.

6.5.3.2 Ensure physically there are no spots on the inner surface.

6.5.4 **Cleaning of Scott bottle and measuring cylinder:**

6.5.4.1 Fill the scott and measuring cylinder up to the over flow level and rinse twice.

6.5.4.2 Ensure physically there are no spots on the inner surface.

6.5.4.3 After completion of individual equipment and accessories cleaning, ensure the system is lined up in the sequence as per the above diagram (required for processing).

6.5.5 **Valve and accessories cleaning:**

6.5.5.1 Before startup of CIP for vessels the valve connected to cooling vessel, Mixing vessel, Holding vessel, buffer tank upstream shall be opened and dipped in to the WFI(collected in Bucket or Bowl) for 5 min two time, reconnect all the valve to the respective area and continue the CIP.

6.5.6 **Cooling vessel and Mixing Vessel:**

6.5.6.1 Ensure the WFI loop circulation pump pressure is NLT 3 Kg/cm^2 before start of cleaning.

6.5.6.2 Connect the cooling vessel to WFI loop through flexible hose pipe on center Tri clover clamp (TC) containing spray ball for cleaning.

6.5.6.3 Open the WFI full valve and allow flushing the vessel inner surface for 5 min. (Approx. 120 L of water flow volume, spray valve will dip and WFI will filled up to the neck.

6.5.6.4 Close the vent and Apply the nitrogen pressure NMT 1kg/cm² and transfer whole the water to the Mixing vessel through connected line (end connection with flexible hose pipe).

6.5.6.5 Disconnect the bottom secondary valve of mixing vessel line to filter housing line and Connect the hose pipe and other end of the hose pipe shall be connected to the drain point.

6.5.6.6 Open the bottom valve and drain the collected WFI, Close the bottom valve after water is drained out.

6.5.6.7 Connect the mixing vessel to WFI loop through flexible hose pipe on center Tri clover clamp (TC) containing spray ball for cleaning.

6.5.6.8 Open the WFI full valve and allow flushing the vessel inner surface for 6 min. (Approx.120 L of water over flow volume, spray valve will dip and WFI will filled up to the neck.)

6.5.6.9 Reconnect the bottom secondary valve of mixing vessel lone to filter housing line.

6.5.6.10 Close the vent and apply the nitrogen pressure NMT 1kg/cm² to mixing vessel.

6.5.6.11 Open the bottom valve (ensure the upstream and downstream valve is remains closed before applying nitrogen pressure)

6.5.6.12 Slowly open the upstream valve and allowed the fill the filter housing and removed the trapped air thorough vent line of filter housing.

6.5.6.13 Open the downstream valve and holding vessel receiving valve and transferred the flushing water to the holding vessel.

6.5.7 **Holding vessel and Buffer tank:**

6.5.7.1 Close the vent and apply the nitrogen pressure NMT 1 Kg/cm² to mixing vessel.

6.5.7.2 Open the bottom valve of holding vessel, buffer tank, drain and Transfer whole the collected water to the drain through buffer tank.

6.5.7.3 Release the nitrogen pressure of holding vessel, closed the bottom valve.

6.5.7.4 Connect the holding vessel to WFI loop through flexible hose pipe on center Tri clover clamp (TC) containing spray ball for cleaning.

6.5.7.5 Open the WFI full valve and allow flushing the vessel inner surface for 6 min. (Approx.120 L of water over flow volume, spray valve will dip and WFI will filled up to the neck.)

6.5.7.6 Close the vent and apply the nitrogen pressure NMT 1 kg/cm² to mixing vessel.

6.5.7.7 Open the bottom valve and transfer the Flushed WFI through Buffer tank through secondary filter housing(release the filter hosing air through vent line to ensure the filled housing is filled housing is filled totally)to drain till vessel gets empty.

6.5.7.8 Disconnect the WFI hose, take out the spray ball piece from the tank and connect it to the buffer vessel.

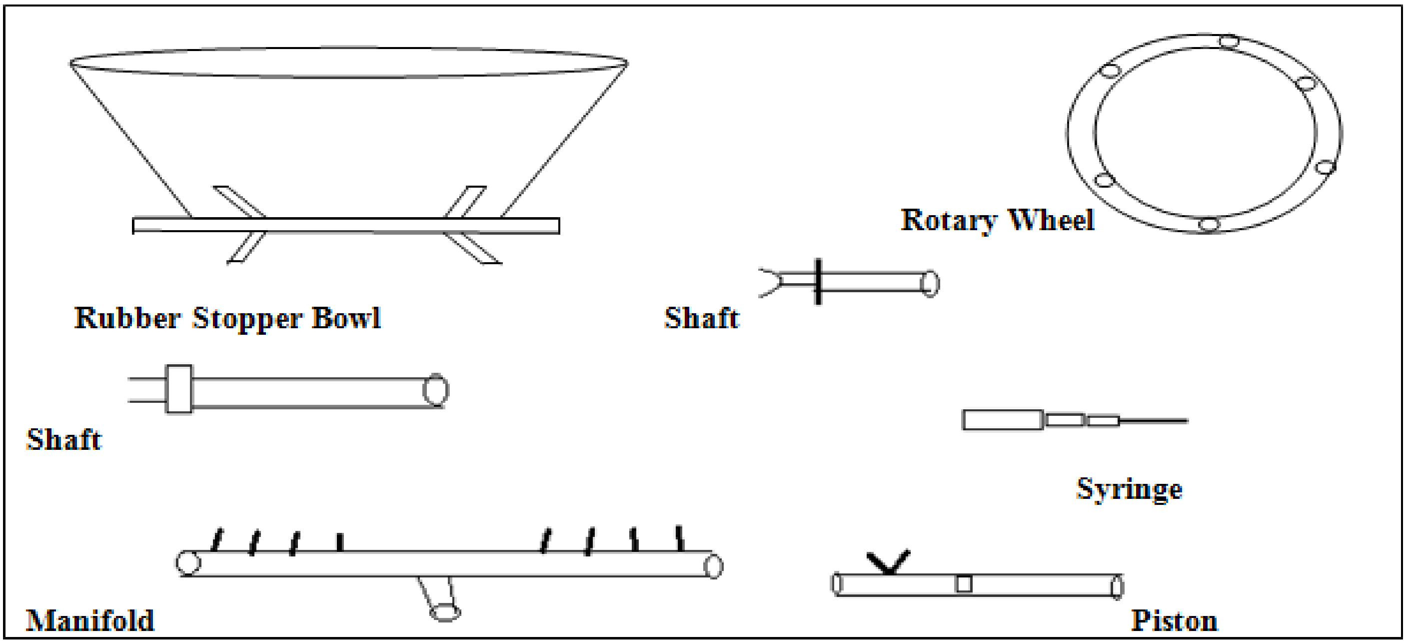
6.5.7.9 Connect the WFI hose to the buffer tank.

6.5.7.10 Open the WFI full valve and allow flushing the buffer tank for 30 sec (5-6lts), keeping drain valve open during flushing.

6.5.8 **After completion of individual equipment and accessories cleaning, ensure the system is lined up in the sequence (required for processing):**

6.5.8.1 Take the WFI in cooling vessel Approx. 120L of water over flow volume, spray valve will dip and WFI will filled up to the neck,
6.5.8.2 Close the vent and apply the nitrogen pressure NMT 1Kg/cm² to mixing vessel.
6.5.8.3 Transfer the WFI to the Mixing vessel.
6.5.8.4 Close the vent and apply the nitrogen pressure NMT 1Kg/cm² to mixing vessel.
6.5.8.5 Open the bottom valve and transfer the mixing vessel WFI to holding vessel followed by primary filter housing (ensure air is released through vent line).
6.5.8.6 After completion of transferring, Close the vent and apply the nitrogen pressure NMT 1Kg/cm² to holding vessel.
6.5.8.7 Open the bottom valve and transfer the holding vessel WFI to the buffer tank followed by secondary filter housing (ensure air is released through vent line).
6.5.8.8 Allow to drain whole the WFI.
6.5.8.9 Rinse sampling.
6.5.8.10 Rinse the Sodium hydroxide vessel with 5L of WFI Collect the rinse sample for the bottom of the vessel.
6.5.8.11 Rinse the cooling vessel with 20L of WFI through spray valve collect the rinse sample from the bottom of the vessel.
6.5.8.12 Drain the remaining water from the cooling vessel.
6.5.8.13 Take again 20L WFI in cooling vessel, close the vent and apply nitrogen pressure to transfer the WFI to the Mixing vessel.
6.5.8.14 Collect the rinse sample from the bottom for the cooling vessel to mixing vessel line flushing.
6.5.8.15 Drain the remaining water from the Mixing vessel.
6.5.8.16 Rinse the mixing vessel with 20 L of WFI through spray valve collect the rinse sample from the bottom of the vessel.
6.5.8.17 Drain the remaining water from the mixing vessel.
6.5.8.18 Take again 20 L WFI in mixing vessel, close the vent and apply nitrogen pressure to transfer the WFI to the holding vessel through primary filter housing(release the filter hosing air through vent line to ensure the filled housing is filled totally).
6.5.8.19 Collect the rinse sample from the bottom for the holding vessel for the line flushing from mixing vessel to holding vessel.
6.5.8.20 Drain the remaining water from the holding vessel.
6.5.8.21 Rinse the holding vessel with 20L of WFI through spray valve collect the rinse sample from the bottom of the vessel.
6.5.8.22 Drain the remaining water from the holding vessel.
6.5.8.23 Take again 20L WFI in holding vessel, close the vent and apply nitrogen pressure to transfer the WFI to the holding vessel through secondary filter housing (release the filter hosing air through vent line to ensure the filled housing is filled totally),open the drain valve and collect the rinse sample from Drain point.
6.5.8.24 Rinse the holding vessel with 5L of WFI through spray valve (ensure spray ball fixed) collect the rinse sample from the bottom of the tank.
6.5.8.25 Drain the remaining water from the holding vessel.

6.5.9 **Cleaning of machine parts:**



6.5.10 Release both the shaft from piston,

6.5.11 Place the, Rotary wheel, rubber stopper chute, Piston 8 no., Pistons shaft 16 no., Syringe 8 no. manifold 1 no, in the washing bin.

6.5.12 Fill the Purified water in the bin to immerse all the parts in the PW.

6.5.13 Hold the accessories in the bin for 10 min.

6.5.14 Drain the water from the bin.

6.5.15 Again filled the bin with PW and hold for 5min.

6.5.16 Drain the water from washing bin, take all the accessories one by one and flushed with PW for8-10sec.

6.5.17 Flushed out the entire washing bin with WFI, put all the accessories in the bin and immersed for 5 min.

6.5.18 Take all the accessories one by one and flashed with WFI for 8-10sec.

6.5.19 Take manifold and connect in 1” TC one end with flexible hose pipe and connect other end of the hose pipe to the WFI Loop line.

6.5.20 Hold the lined up manifold in the washing bin.

6.5.21 Open the WFI loop valve and allowed to flush the manifold (containing 8 no. nozzles) till washing bin gets filled.

6.5.22 Drain the flushed water of washing bin and again flush the manifold till the washing bin gets full.

6.5.23 Take new silicon tubes 8 no. cut in to the pieces (NMT 1 Meter) connect one end of the silicon tube to the manifold nozzle(8no.) and one end to the syringe, 6.5.24 Flushed the connected system with WFI and collected 10 L WFI, withdraw the rinse sample for analysis as per the SOP.

6.5.25 Take 10 L WFI in a bucket, dip the Piston and shaft for 5 min. and withdraw the rinse sample for analysis as per the SOP.

6.5.26 Flushed the external surface of the rubber stopper bowl with Purified water for 8-10 sec.

6.5.27 Filled the rubber stopper bowl with purified water up to over flow level and hold for 5 min,

6.5.28 Drain the Purified water to drain.

6.5.29 Flushed the external surface of the rubber stopper bowl with WFI for 8-10sec.

6.5.30 Filled the rubber stopper bowl with purified water up to over flow level and hold for 5 min,

6.5.31 Drain the Purified water to drain.

6.5.32 Take 5L of WFI in the bowl put rotary wheel and rubber stopper chute for rinse sample hold for 5 min, withdraw the rinse sample for analysis as per the SOP.

6.5.33 **Lyophilizer:**

6.5.33.1 Lyophilizer is cleaned by Automated CIP system and during final cleaning withdraws the rinse sample for analysis as per the SOP.

6.6 Rinse samples shall be collected from defined locations as mentioned in **Annexure – 01** and Send all samples along with blank sample to QC department for analysis of following test:

- Visual particles
- Sub visual particles
- Residue of previous product
- BET
- Bio burden

6.7 Depyrogenated container shall be used for BET sample collection.

6.7.1 Use the pre validated analytical method for analysis.

☞ *If samples are not analyzed immediately then samples are stores at 2 to 8°C and Samples shall be analyzed within 24 hours for chemical analysis.*

☞ *If samples are not analyzed immediately then samples are stores at 2 to 8°C and Samples shall be analyzed within 12 hours for microbial analysis.*

☞ *For bio burden sample use autoclaved bottle.*

6.8 **Recovery, LOQ and LOD Study:**

Recovery study, LOQ and LOD study is performed as per the analytical cleaning method validation protocol as follows -.

6.8.1 Analytical Method Validation protocol for Ertapenem for Injection by HPLC.

6.8.2 **Testing Plan:**

6.8.2.1 **Rinse sample:** Visually inspect to particulate matter and analyze the rinse sample by suitable HPLC method for residue of previous product in the sample. BET and bio burden to be analyzed by suitable method. Use the pre validated analytical method for analysis.

☞ *Particulate matters to be check firstly, if it is complying then proceed for next analysis.*

7. ACCEPTANCE CRITERIA:

7.1 Should be visually clean.

7.2 Should be free from visible particle and sub visible particle ≥ 10µ: NMT 25/ml
≥ 25µ: NMT 3/ml

7.3 Residue of previous product in next product should be not more than 10 ppm (0.001%).

7.4 For the product non-contact surfaces such as external surface of equipment etc, an acceptance chemical residue limit of 100 PPM per swab shall be considered.

7.5 The acceptance limit for microbial load is as follows:

- Bacterial Endotoxin Test shall be < 0.125 EU/ml.
- Bio burden shall be less than < 10 CFU / 100ml.

8. REVALIDATION CRITERIA:

Process to be revalidated in any of the following case:

8.1 Equipment change with different design specification.

8.2 Major modifications / changes to the equipments.

8.3 Change in the cleaning procedure.

8.4 Change in the manufacturing process.

8.5 Addition of new molecule.

8.6 MACO value decreased.

8.7 **Periodic verification:**

8.7.1 Periodic verification shall be done once in a year (± 30 days) with product, precisely the worst case product to cover all the equipments for those equipment on which worst case product is not taken, consider the worst case product from the group of products sharing that equipment.

☞ *Periodic verification shall be carried out as per approved protocol.*

9. OBSERVED NON-CONFORMANCE (IF ANY):

All the non-conformance shall be addressed as per “Reporting and Monitoring of Process Non-Conformance in the Automated Quality Management System Software”.

10. VALIDATION REPORT:

On completion of the cleaning validation, evaluation of the test results shall be carried out for achievement of the acceptance criteria. A summary report shall be prepared clearly stating the outcome of the cleaning validation against the predetermined acceptance criteria.

11. ABBREVIATIONS:

11.1 MACO : Maximum Allowable Carry Over
11.2 MAR : Maximum Allowable Residue
11.3 HPLC : High Performance Liquid Chromatography
11.4 NMT : Not More Than
11.5 ppm : Parts Per Million
11.6 BET : Bacterial Endotoxin Test
11.7 TDD : Therapeutic Daily Dose
11.8 MBS : Minimum Batch Size
11.9 SF : Safety Factor
11.10 cm^2 : Centimeter square
11.11 CFU : Colony Forming Unit
11.12 EU : Endotoxin Unit

12. LIST OF ANNEXURE:

Annexure No.	Annexure Title
01	Rinse Sample location
02	Visual inspection for cleaning validation / verification
03	Equipment / equipment parts cleaning record
04	Equipment rinse sample analysis test results
05	Details of sampling and visual inspection observation

13. REFERENCE DOCUMENT (IF ANY):

13.1 Management of Validation/Qualification documents
13.2 Cleaning Validation Programme
13.3 Preparation of Validation and Qualification Protocol and its Control

14. REVISION HISTORY:

Superseded Protocol		S. No.	Step No.	Changes made
Protocol No. / Version No.	Effective Date			

Annexure – 01: Rinse sample locations

S.No.	Equipment Name	Sampling Location
1	NaOH cooling vessel	Vessels to be flushed with 5 L of WFI and withdraw the sample from bottom of the vessel.
2	Cooling vessel	Vessels to be flushed with 20 L of WFI and withdraw the sample from bottom of the vessel.
3	Transfer line-Cooling vessel to Mixing vessel	Collect 20 L WFI in the cooling vessel transfers the WFI through transferring line and withdraw the sample from bottom of the vessel.
4	Mixing vessel	Vessels to be flushed with 20 L of WFI and withdraw the sample from bottom of the vessel.
5	Holding vessel	Vessels to be flushed with 20 L of WFI and withdraw the sample from bottom of the vessel.
6	Transfer line from mixing vessel to holding vessel through Filtration train-1	Collect 20 L WFI in the cooling vessel transfer the WFI through transferring line and withdraw the sample from bottom of the vessel.
7	Transfer line from Holding vessel to buffer tank and Filling machine transfer line through Filtration train -2	Collect 20 L WFI in the cooling vessel transfer the WFI through transferring line and withdraw the sample from bottom of the vessel.
8	Buffer Tank	To be flushed with 5 L of WFI and withdraw the sample from drain line.
9	Piston, Shaft	Sample to be collected from bucket
10	Syringe with Manifold	Sample to be collected during flushing of the system.
11	Rubber stopper Bowl, rubber stopper chute, rotary wheel	Sample to be collected from bucket.

Annexure – 02: Visual inspection for cleaning validation/verification

S.No.	Product Name/ Batch No.	Equipment Name	Equipment ID No.	Part Inspected	Visually clean (Yes/No)	Checked by Sign/Date

Annexure – 03: Equipment / equipment parts cleaning record

S.No.	Product Name/ Batch No.	Equipment /equipment part name	Equipment ID No.	WFI loop pressure	Cleaning		Cleaning done by (Sign/Date)	Checked by Sign/Date
					From	To		

Annexure – 04: Equipment rinse sample analysis test results

Date :

Product Name :

Batch No :

S. No.	Sample Description	Observation						Test results (Complies/ Not Complies)	Complied By	Verified By	Remarks
		BET		Particulate matter		TVAC					
		Test Date	Results	Test Date	Results	Test Date	Results				
1	NaOH cooling vessel										
2	Cooling vessel										
3	Transfer line Cooling vessel to Mixing vessel										
4	Mixing vessel										
5	Holding vessel										
6	Transfer line from mixing vessel to holding vessel through Filtration train-1										
7.	Transfer line from Holding vessel to Buffer tank and filling machine transfer line through Filtration train-2										
8	Buffer tank										
9	Piston, Shaft										
10	Syringe with Manifold										
11	Rubber										

S. No.	Sample Description	Observation						Test results (Complies/ Not Complies)	Complied By	Verified By	Remarks
		BET		Particulate matter		TVAC					
		Test Date	Results	Test Date	Results	Test Date	Results				
	stopper Bowl, rubber Stopper chute, rotary wheel										

Reviewed By:
(Sign/Date)

PHARMA DEVILS	PROTOCOL FOR CLEANING VALIDATION - PRODUCTION LYOPHILIZATION	PROTOCOL No.:

Annexure – 05: Details of sampling and visual inspection observation

Date :

Product Name :

Batch No :

S. No.	Sample Description	Date of equipment cleaning	Equipment surface visually cleaned (Y/N)		Rinse sample				Remarks
			Observation	Checked by	Sampling		Rinse sample visual inspection for particulate matter		
					Qty.	Done by	Observation	Checked by	
1	NaOH cooling vessel								
2	Cooling vessel								
3	Transfer line Cooling vessel to Mixing vessel								
4	Mixing vessel								
5	Holding vessel								
6	Transfer line from mixing vessel to holding vessel through Filtration train-1								
7.	Transfer line from Holding vessel to Buffer tank and filling machine transfer line through Filtration train-2								
8	Buffer tank								
9	Piston, Shaft								
10	Syringe with Manifold								
11	Rubber stopper bowl, rubber stopper chute, rotary wheel								

Reviewed By:
(Sign/Date)