

(1) **Validation of Protocol and Data**

TEST	DOCUMENTS REQUIRED	AVAILABILITY
STERILITY TEST (ST)	1. CoA with specification and result for ST	
	2. List of media and reagent a. Culture media b. List of rinsing solution, buffer solution and diluents c. Neutralizing agent (if any)	
	3. Preparation of media & Composition of rinsing buffer	
	4. Method of Test for Media Sterility and Growth Promotion Test	
	5. Preparation of test sample (including steps to eliminate antimicrobial activity due to antibiotic samples or samples which contain preservatives).	
	6. Detailed test procedure for sterility test a. Quantity of sample/ Volume of sample b. Membrane filtration or direct inoculation c. Open System or Closed System (if Membrane filtration method used) d. Volume of rinsing fluid used for each membrane	
	7. Validation data for Sterility Test (Bacteriostasis dan fungistatsis test) a. Quantity of sample/ Volume of sample b. Membrane filtration or direct inoculation c. Open System or Closed System (if Membrane filtration method used) d. Volume of rinsing fluid used for each membrane Do all the details above are same as sterility test?	
	8. Result (raw data) - with Name & Strength of product, Batch No of product, date of test started and ended, observation in every interval period a. Test for Media Sterility and Growth Promotion Test b. Sterility Test c. Validation for sterility test (Bacteriostasis dan fungistatsis test)	
	9. Does reconstitute solution provided? If yes, method and result of sterility test & validation test should be provided.	

MICROBIAL CONTAMINATION TEST (MCT)	1. CoA with specification and result for MCT	
	2. Specification and acceptance criteria	
	3. Preparation of media	
	4. Test for Growth Promoting and Inhibitory Properties of Media & Media Sterility Test	
	5. Preparation of test sample (including neutralizing of preservatives for samples that contain preservatives)	
	6. Method of MCT (Routine Test) <ul style="list-style-type: none"> a. Total Viable Aerobic Count <ul style="list-style-type: none"> ➤ Detailed test procedure for Total Aerobic Microbial Count (TAMC) and Total Yeasts and Moulds Count (TYMC) - by Plate Count, Membrane Filtration or Most-Probable Number (MPN) method. b. Test for Specified Microorganisms <ul style="list-style-type: none"> ➤ Detailed test procedure for each specific microorganism tested (including identification and confirmation test) 	
	7. Validation data for MCT <ul style="list-style-type: none"> a. Total Viable Aerobic Count (Suitability of the counting method in the presence of product) <ul style="list-style-type: none"> ➤ Acceptance criteria: Mean count of any test organisms not differing by a factor greater than 2 (50% – 200%) b. Test for Specified Microorganisms (Suitability of the test method) <ul style="list-style-type: none"> ➤ Spiked microorganisms must be detected <p>Do all the details of testing procedure same as routine test?</p>	
	8. Result (raw data) - with Name & Strength of product, Batch No of product, date of test started and ended, observation in every interval period <ul style="list-style-type: none"> a. Test for Growth Promoting and Inhibitory Properties of Media & Media Sterility Test b. MCT (Routine Test) <ul style="list-style-type: none"> ➤ Total Viable Aerobic Count (TAMC & TYMC) ➤ Test for Specified Microorganisms c. Validation Test result <ul style="list-style-type: none"> ➤ Total Viable Aerobic Count (TAMC & TYMC) ➤ Test for Specified Microorganisms 	

Biological Assay of Antibiotics	<p>1. Procedure for preparation of following solutions/ substances:</p> <ul style="list-style-type: none"> • Culture medium • Buffer solutions • Diluents • Microorganisms used in assay 	
	<p>2. Detailed test method (diffusion or turbidimetric method), which includes:</p> <ul style="list-style-type: none"> • Preparation of standard solutions (including steps to counteract the antimicrobial properties of any preservatives, etc present in the sample) • Preparation of test solutions (including any steps to neutralize the antimicrobial properties of any preservatives, etc present in the sample) • Test for Media Sterility and Growth Promotion Test • Dilution schemes for test and standard solutions <ul style="list-style-type: none"> i. Application of test & standard solutions (volume, use of latin squares, etc) ii. Incubation temperature & time iii. Interpretation of result iv. Detailed calculation for the test including ANOVA table and other data showing validity of test results 	