STANDARD OPERATING PROCEDURE

Title: Preparation Storage and Use of Culture Media				
SOP No.:		**Department:**	Microbiology	
		Effective Date:		
Revision No.:	00	**Revision Date:**		
Supersede Revision No.:	Nil	**Page No.:**	1 of 20	

1.0 OBJECTIVE

To lay down procedure for preparation, storage and use of culture media for microbiological analysis.

2.0 SCOPE

This SOP is applicable for preparation, storage and use of culture media for microbiological analysis carried out in Microbiology laboratory.

3.0 RESPONSIBILITY

Execution - Executive Microbiology

Checking- Assistant Manager Microbiology

Overall Compliance – Head QA, QC

4.0 PROCEDURE

4.1 Receiving, Storage and Usage of Dehydrated Media

4.1.1 After receiving the material check that the material received is in proper condition or not.

4.1.2 Check that the material condition is satisfactory or not.

4.1.3 Check the manufacturing date, expiry date, lot number etc.

4.1.4 Reject the material if any defects are observed in received condition.

4.1.5 Place a label on the dehydrated culture media bottle.

4.1.6 Labels must contains the following details as per Annexure - I.

4.1.7 Enter the receipt details in the stock record as per Annexure - II.

4.1.8 In case of newly received lots of dehydrated media check the growth promoting ability of the media.

4.1.9 If the growth promotion test results are found satisfactory accept the media.

4.1.10 If the growth promotion test results are found not satisfactory reject the media.

4.1.11 Store the dehydrated media at temperature NMT 25^0C or at prescribed storage temperature or suggested by the manufacturer.

4.1.12 Follow FEFO (First Expiry First Out) and consume all the dehydrated media.

4.1.13 Use the dehydrated media Up to expiry date as per given by Manufacturer.

4.2 Media Preparation

4.2.1 After receiving the material check that the material received is in proper condition or not.

4.2.2 Preferably use the pre-formulated dehydrated media for media preparation.

4.2.3 If pre-formulated dehydrated media is not available, prepare the media from its basic ingredients as per direction of pharmacopoeia or respective test procedure.

4.2.4 Weigh the required quantity of the dehydrated media in a suitable container as per the manufacturer's instruction and dissolve it in Water for injection/water for injection and make up the specified volume with purified water/water for injection.

4.2.5 Dissolve the media by shaking, tilting and /or heating with continuous stirring, if required.

4.2.6 Measure the pH by using a calibrated pH meter and adjust the pH with 0.1N HCl /0.1N NaOH as required.

4.2.7 Distribute the required volume of media or reagents etc. in appropriate glass containers.

4.2.8 Volume of media in each container should not be more then 1/3 capacity of the container.

4.2.9 Allocate the lot no. to each prepared media as following.

XXXX	B.No.	YY / ZZZZ

Where XXXX is the short form represents the media name in capital alphabets. Batch No. is the Manufacturer Batch No./Lot No., if the media is prepared by its basic ingredient then writes the short form of the reference document in capital alpha bets from which the composition referred BP for British Pharmacopoeia, USP for United State Pharmacopoeia, IP for Indian Pharmacopoeia, EP for European Pharmacopoeia and IH (In House) for other than any Pharmacopoeia.

YY represents the year in which sterilization carried out.

ZZZZ is denoting the autoclave load no. in which the particular media to be sterilised.

For example - Lot no. of the media

SCDA	2J6656	23 / 0001

Soyabean Casein Digest agar Media of Batch No. 2J6656 with autoclave load no. 0001 of the year 23.

4.2.10 Paste the label indicating the assigned Lot no., and details on to the each media container as per Annexure - III.

4.2.11 Close the media container by cotton plug and cover with butter paper or caps and sterilize as per specified instructions.

4.2.11 For steam sterilization place the media container in autoclave as per SOP.

4.2.12 After sterilization, cool down the liquid media at room temperature.

4.2.13 In case of solid media, cool it up to 45 - 50 ºC with ensuring the molten form and distribute.

4.2.14 If any delay in distribution then to maintain its molten form keep it on to a hot plate or oven or water bath at a temperature 45 - 50 ºC up to a period NMT 6 Hrs.

4.2.15 **Slant Preparation -** Fill the agar media up to ¼ spaces of test tubes and after sterilization keep the tubes at approximately 20 to 30 º angle position to form slant and allow solidifying. After solidification preincubate the agar slants as per Table - III.

4.2.16 **Stab Preparation -** Fill the agar media up to 1/ 2 space of test tubes and after sterilization keep the tubes in the upright position to form the stab and allow for solidifying. After solidification preincubate the agar stabs as per Table - III.

4.2.17 **Plate Preparation for Settle Plate Exposure -** After sterilization aseptically pour 15 to 20 ml of molten agar media in sterile 90mm petri plate and keep in the upright position and allow it to solidify. After solidification preincubate the agar plates as per Table - III.

4.2.18 **Contact Plate Preparation -** After sterilization aseptically pour molten agar media in the contact plates or RODAC plates in such a way that the surface of the medium is slightly raised in comparison to the edge of the plate, cover the lid of the plate and allow them for solidify. After solidification preincubate the agar plates as per Table - III.

4.2.19 **Active Air Sampling Plates Preparation -** After sterilization aseptically pour sufficient quantity of molten agar media in to the empty cassettes (Air sampling plate), cover the lid of the plate and allow them for solidify. After solidification preincubate the agar plates as per Table - III.

4.2.19 **Preparation of Swabs for Surface Monitoring -** Put the SWAB sticks individually in test

tubes containing 5 ml 0.9 % normal saline or buffered peptone water and plug the test tubes with PP caps or cotton plugs or transfer 5 ml 0.9 % normal saline or buffered peptone water in ready to used swabs and again closed the swabs. Keep the tubes/swabs in a stand, wrap the stand with butter paper and sterilize by autoclaving at as per SOP.

4.2.21 In case of solid media not used on the day of preparation store the media below 25^0C and before use melt it by boiling only once “Do not autoclave and over heat” cool it up to 45 - 50 ºC with ensuring the molten form and distribute.

4.2.22 **Preparation of Thermo Labile Media using Boiling Method -** Weigh the required quantity of the media and prepare it as per Table - I.

4.2.23 Maintain the media preparation record as per Annexure - IV and V.

Table - I

Preparation of Thermo Labile Media

Media	Quantity in gm/liter	Method for preparation of media
Bismuth Sulphite Agar	As per manufacturer instruction	Heat to boiling to dissolve the media completely in Water for injection / water for injection. Cool to 50 - 55º C. Mix well to disperse precipitated bismuth Sulphite and pour into sterilized petri plates.
Selenite F - broth		Dissolve the media completely in purified water / water for injection. Dispense in sterilized test tubes. Sterilize in boiling water bath for 30 minutes.
Tetrathionate Brilliant Green Bile Broth		Dissolve the media in Water for injection / water for injection and heat till the media boils. Add freshly prepared solution containing 5 g potassium iodide and 6 g iodine dissolved in 20 ml. Sterilized distilled water into the medium before use. Dispense the medium in sterilized test tubes.
Tetra Thionate Broth Base		Dissolve the media in sterilized Water for injection / water for injection and heat till the media boils. Add freshly prepared solution containing 5 g potassium iodide and 6 g iodine dissolved in 20 ml. Sterilized distilled water and 10 ml of 0.1% brillant green solution into the medium before use. Dispense the medium in sterilized test tubes.
Deoxycholate Citrate Agar		Heat to boiling to dissolve the media completely in Water for injection / water for injection. Cool to 50 - 55º C. Mix well pour into sterilized petri plates.

Table - I

Preparation of Thermo Labile Media

Media	Quantity in gm/liter	Method for preparation of media
Xylose Lysine Deoxycholate Agar		Heat with frequent agitation until the medium boils. Cool to 50 - 55º C. Mix well pour into sterilized petri plates.
Enterobacteriaceae enrichment broth mossel		Dispense the medium in sterilized test tubes or flasks. Dissolve the media in Water for injection and boil on water bath for 30 minutes.
Fluid Selenite Cystine Medium		Dissolve the media in Water for injection and heat till the media boils. Dispense the medium in sterilized test tubes

4.3 Storage of Prepared Media

Store all the prepared media e.g. plates, broth tubes, slants, stabs and broth or solid media container at a validated temperature for validated time.

4.4 Preparation of 10 - 100 cfu Culture Suspension

4.4.1 Take out the working cultures from the refrigerator 30 minutes prior to the testing so as to acclimatize with the working environment and place it under laminar airflow unit.

4.4.2 Red hot the loop and cool it, take a loop full of culture and inoculate it into 50 ml of sterile 0.9 % saline solution (initial suspension).

4.4.3 Gently vortex it so as to make a uniform suspension.

4.4.4 With the help of micropipette take 5.0 ml from the Initial suspension and inoculate it into 45 ml of 0.9 % saline solution (1:10 dilution).

4.4.5 Transfer 5.0 ml from the above dilution to another tube containing 45 ml of the 0.9 % saline solution.

4.4.6 Perform the serial dilutions in the same manner ranging from 1:10 to1: 100000000.

4.4.7 Perform the whole exercise with all the organisms, which are being used for growth promotion test.

4.4.8 Enumerate the culture organism by pour plate method and add 1.0 ml of the dilutions into sterile Petri plates.

4.4.9 Pour sterile molten Soyabean casein digest agar media to all the plates containing Respective dilution for bacterial cultures and Sabourauds dextrose agar media to all the yeast, fungal or molds cultures into the plates.

4.4.10 Incubate anaerobic culture in anaerobic jar for at 32.5 ± 2.5 ° C for 72 hrs.

4.4.11 Incubate the plates at 32.5 ± 2.5°C for 48 - 72 hrs for bacterial cultures and 22.5 ± 2.5°C for 72 - 120 hrs for fungi.

4.4.11 Do not discard the dilutions after plating the cultures. Preserve the dilutions in refrigerator at temperature 2-8°C.

4.4.12 After the specified incubation period enumerate the number of colonies.

4.4.13 Select the dilution which is having cells between 10 - 100 cfu / ml.

4.4.14 Preserve the dilution which is having cells between 10 - 100 cfu / ml in refrigerator for further tests.

4.4.15 Preserve the anaerobic culture suspension by adding 3 ml of mineral oil to prevent air contact with the suspension.

4.4.16 Store the culture suspension for one week at 2-8°C.

4.4.17 Frequency of bacterial and fungal culture suspension preparation is once in a week.

4.4.18 Label the culture suspension with appropriate details as per annexure - VI.

4.4.19 Maintain the culture suspension preparation record as per annexure - VII.

4.4.20 Decontaminate and discard the left over dilutions by decontamination procedure as per SOP.

4.5 Growth Promotion Test

4.5.1 Perform the growth promoting properties of the each autoclave lot of medium using NMT 100 cfu of challenge organism as specified in Table - II.

4.5.2 Perform the growth promotion test in duplicate.

4.5.3 Incubation is to be carried out as specified in Table - II.

4.5.4 The medium is satisfactory for use, if growth observed with in 48 hrs for bacteria and with in 120 hrs for fungi as per the specific incubation condition.

Table - II
Growth Promotion Test of Media

Media	Abbreviations	Challenge organisms for growth promotion test		Incubation condition for growth promotion test	
		Name	MTCC No.	Time (hrs)	Temperature
Soyabean Casein Digest Agar	SCDA **	*A.niger*	MTCC 1344	72-120	20 - 25 °C
		C. albicans	MTCC 227	72-120	20 - 25 °C
		S. aureus	MTCC 737	48 - 72	30 - 35 °C
		B. subtilis	MTCC 441	48 - 72	30 - 35 °C
		P. aeruginosa	MTCC 1688	48 - 72	30 - 35 °C
		E. coli	MTCC 1687	48 - 72	30 - 35 °C
Sabourauds Dextrose Agar	SDA	*A.niger*	MTCC 1344	72-120	20 - 25 °C
		C. albicans	MTCC 227	72-120	20 - 25 °C
Sabourauds Chloramphenicol Agar	SCA	*A.niger*	MTCC 1344	72-120	20 - 25 °C
		C. albicans	MTCC 227	72-120	20 - 25 °C
Potato Dextrose Agar	PDA	*A.niger*	MTCC 1344	72-120	20 - 25 °C
		C. albicans	MTCC 227	72-120	20 - 25 °C
Soyabean Casein Digest Medium	SCDM **	*A.niger*	MTCC 1344	72-120	20 - 25 °C
		C. albicans	MTCC 227	72-120	20 - 25 °C
		S. aureus	MTCC 737	48 - 72	30 - 35 °C
		B. subtilis	MTCC 441	48 - 72	30 - 35 °C
		P. aeruginosa	MTCC 1688	48 - 72	30 - 35 °C
		E. coli	MTCC 1687	48 - 72	30 - 35 °C
Xylose Lysine Deoxycholate Agar	XLDA	*Salmonella spp.*	MTCC 3858	48 - 72	30 - 35 °C
Peptone Water	PW	*E. coli*	MTCC 1687	48 - 72	30 - 35 °C
Bacteriological Peptone	BP	NA	NA	NA	NA
Bismuth Sulphite Agar	BSA	*Salmonella spp.*	MTCC 3858	48 - 72	30 - 35 °C
Mac Conkey Agar	MA	*E. coli*	MTCC 1687	48 - 72	30 - 35 °C
Mannitol Salt Agar	MSA	*S. aureus*	MTCC 737	48 - 72	30 - 35 °C
Cetrimide Agar	CA	*P. aeruginosa*	MTCC 1688	48 - 72	30 - 35 °C
Buffered Peptone Water	BPW	NA	NA	NA	NA
Vogel Johnson Agar	VJA	*S. aureus*	MTCC 737	48 - 72	30 - 35 °C
Tetra Thionate Broth Base	TTBB	*Salmonella spp.*	MTCC 3858	48 - 72	30 - 35 °C

Table - II
Growth Promotion Test of Media

Media	Abbreviations	Challenge organisms for growth promotion test		Incubation condition for growth promotion test	
		Name	MTCC No.	Time (hrs)	Temperature
Nutrient Broth	NB **	*A.niger*	MTCC 1344	72-120	20 - 25 °C
		C. albicans	MTCC 227	72-120	20 - 25 °C
		S. aureus	MTCC 737	48 - 72	30 - 35 °C
		B. subtilis	MTCC 441	48 - 72	30 - 35 °C
		P. aeruginosa	MTCC 1688	48 - 72	30 - 35 °C
		E. coli	MTCC 1687	48 - 72	30 - 35 °C
Fluid Lactose Medium	FLM **	*A.niger*	MTCC 1344	72-120	20 - 25 °C
		C. albicans	MTCC 227	72-120	20 - 25 °C
		S. aureus	MTCC 737	48 - 72	30 - 35 °C
		B. subtilis	MTCC 441	48 - 72	30 - 35 °C
		P. aeruginosa	MTCC 1688	48 - 72	30 - 35 °C
		E. coli	MTCC 1687	48 - 72	30 - 35 °C
Eosin Methylene Blue Agar	EMBA	*E. coli*	MTCC 1687	48 - 72	30 - 35 °C
Fluid Selenite Cystine Medium	FSCM	*Salmonella spp.*	MTCC 3858	48 - 72	30 - 35 °C
Triple Sugar Iron Agar	TSIA	*Salmonella spp.*	MTCC 3858	48 - 72	30 - 35 °C
Mac Conkey Broth purple	MB	*E. coli*	MTCC 1687	48 - 72	30 - 35 °C
Baird Parker Agar	BPA	*S. aureus*	MTCC 737	48 - 72	30 - 35 °C
Tetrathionate Brilliant Green Bile Broth	TBGBB	*Salmonella spp.*	MTCC 3858	48 - 72	30 - 35 °C
Fluid Thioglycollate Medium	FTGM	*S. aureus*	MTCC 737	48 - 72	30 - 35 °C
		P. aeruginosa	MTCC 1688	48 - 72	30 - 35 °C
		C.sporogenes	MTCC 1349	48 - 72	30 - 35 °C
Dey Englay Neutrilizing Broth / Dey Englay Neutrilizing Agar	DENB / DENA	*E. coli*	MTCC 1687	48 - 72	30 - 35 °C
		Salmonella spp.	MTCC 3858	48 - 72	30 - 35 °C
		P. aeruginosa	MTCC 1688	48 - 72	30 - 35 °C
		S. aureus	MTCC 737	48 - 72	30 - 35 °C
		B. subtilis	MTCC 441	48 - 72	30 - 35 °C
		A.niger	MTCC 1344	72-120	20 - 25 °C
		C. albicans	MTCC 227	72-120	20 - 25 °C

* * - Any two bacterial /fungal challenge organisms can be used for growth promotion test of the medium.

Table - II
Growth Promotion Test of Media

Media	Abbreviations	Challenge organisms for growth promotion test		Incubation condition for growth promotion test	
		Name	MTCC No.	Time (hrs)	Temperature
Pseudomonas agar (Fluorescence)	PAF	*P. aeruginosa*	MTCC 1688	48 - 72	30 - 35°C
Pseudomonas Agar (Pycocyanin)	PAP	*P. aeruginosa*	MTCC 1688	48 - 72	30 - 35°C
Enterobacteriaceae enrichment broth mossel	EEBM	*E. coli*	MTCC 1687	48 - 72	30 - 35°C
Cooked Meat Medium	CMM	*C.sporogenes*	MTCC 1349	48 - 72	30 - 35°C
Brilliant Green Agar	BGA	*Salmonella spp.*	MTCC 3858	48 - 72	30 - 35°C
Selenite F - broth	SFB	*Salmonella spp.*	MTCC 3858	48 - 72	30 - 35°C
Deoxycholate Citrate Agar	DCA	*Salmonella spp.*	MTCC 3858	48 - 72	30 - 35°C
R2A Agar	R2AA **	*A.niger*	MTCC 1344	72-120	20 - 25°C
		C. albicans	MTCC 227	72-120	20 - 25°C
		S. aureus	MTCC 737	48 - 72	30 - 35°C
		B. subtilis	MTCC 441	48 - 72	30 - 35°C
		P. aeruginosa	MTCC 1688	48 - 72	30 - 35°C
		E. coli	MTCC 1687	48 - 72	30 - 35°C
Plate Count Agar	PCA **	*A.niger*	MTCC 1344	72-120	20 - 25°C
		C. albicans	MTCC 227	72-120	20 - 25°C
		S. aureus	MTCC 737	48 - 72	30 - 35°C
		B. subtilis	MTCC 441	48 - 72	30 - 35°C
		P. aeruginosa	MTCC 1688	48 - 72	30 - 35°C
		E. coli	MTCC 1687	48 - 72	30 - 35°C
Nutrient Agar	NA **	*A.niger*	MTCC 1344	72-120	20 - 25°C
		C. albicans	MTCC 227	72-120	20 - 25°C
		S. aureus	MTCC 737	48 - 72	30 - 35°C
		B. subtilis	MTCC 441	48 - 72	30 - 35°C
		P. aeruginosa	MTCC 1688	48 - 72	30 - 35°C
		E. coli	MTCC 1687	48 - 72	30 - 35°C

* * - Any two bacterial /fungal challenge organisms can be used for growth promotion test of the medium.

4.5 Pre incubation Test

Pre incubate the agar media slants, stab, settle plates, contact plates and air sampling plates as specified in Table - III.

Table - III
Preincubation Test of Media

Media	Abbreviations	Incubation condition for Pre incubation test	
		Time (hrs)	Temperature
Potato Dextrose Agar	PDA	24 - 48 hrs	20 - 25°C
Sabourauds Chloramphenicol Agar	SCA		20 - 25°C
Mac Conkey Agar	MA		30 - 35°C
Mannitol Salt Agar	MSA		30 - 35°C
Vogel Johnson Agar	VJA		30 - 35°C
Sabourauds Dextrose Agar	SDA		20 - 25°C
Baird Parker Agar	BPA		30 - 35°C
Eosin Methylene Blue Agar	EMBA		30 - 35°C
Xylose Lysine Deoxycholate Agar	XLDA		30 - 35°C
Brilliant Green Agar	BGA		30 - 35°C
Deoxycholate Citrate Agar	DCA		30 - 35°C
Pseudomonas agar (Fluorescence)	PAF		30 - 35°C
Pseudomonas Agar (Pycocyanin)	PAP		30 - 35°C
R2A Agar	R2AA		30 - 35°C
Cetrimide Agar	CA		30 - 35°C
Nutrient Agar	NA		30 - 35°C
Plate Count Agar	PCA		30 - 35°C
Triple Sugar Iron Agar	TSIA		30 - 35°C
Bismuth Sulphite Agar	BSA		30 - 35°C
Soyabean Casein Digest Agar	SCDA		30 - 35°C
Dey Englay Neutrilizing Agar	DENA		30 - 35°C

5.0 SAFETY & PRECAUTIONS

Not Applicable

6.0 REVISION HISTORY

Revision No.	Reason for Revision	Superseded from & date
00	First Issue	--------

7.0 REFERENCES

SOP .

8.0 ABBREVIATIONS

SOP	:	Standard Operating Procedure
mL	:	Milliliter
°C	:	Degree Centigrade
NLT	:	Not less than
NMT	:	Not more than
CFU	:	Colony forming unit
Hrs	:	Hours

9.0 ANNEXURES

Annexure - I	**:**	Dehydrated media label
Annexure - II	**:**	Dehydrated media stock record
Annexure - III	**:**	Media label for sterilization
Annexure - IV	**:**	Media preparation and usage record - I
Annexure - V	**:**	Media preparation and usage record - II
Annexure - VI	**:**	Culture dilution / suspension label
Annexure - VII:		Culture suspension preparation record

ANNEXURE - I
DEHYDRATED MEDIA LABEL

Dehydrated Media Label

Date of receipt: _____

Sign: _____

Opening date: _____ Sign: _____

Release date: _____ Sign: _____

Use Before date: _____ Sign: _____

STANDARD OPERATING PROCEDURE

Title: Preparation Storage and Use of Culture Media				
SOP No.:		**Department:**	Microbiology	
		Effective Date:		
Revision No.:	00	**Revision Date:**		
Supersede Revision No.:	Nil	**Page No.:**	13 of 20	

ANNEXURE - II
DEHYDRATED MEDIA STOCK RECORD

Name of the Medium / Code No.: _____ Make: _____

Date	Opening Balance	Details of Receipts			Details of Issue		Closing Balance	Remarks	Sign	Checked by
		Received Quantity	Batch/ Lot No.	Expiry Date	Issue Quantity	Batch/ Lot No.				

ANNEXURE - III
MEDIA LABEL FOR STERILIZATION

Media Label For Sterilization

Name of Medium: _____

Autoclave Load No.: _____

Quantity Dispensed: _____

Prepared and Sterilization On: _____

Use Before date: _____ Sign: _____

ANNEXURE - IV
MEDIA PREPARATION AND USAGE RECORD - I

Name of the Medium -	Date of Preparation -
Name of the Manufacturer -	Manufacturer Lot No. -
Medium to be used for	

Dehydrated medium powder characteristics - Free flowing, Uniform colour, Free from and lumps - are Satisfactory					YES / NO
Quantity of powder weighed (g)			Volume of water for injection added (ml)		
Analytical Balance ID			Boiling of medium performed		YES / NO
pH Meter ID					
pH before sterilization		pH adjusted to (if any)		*Medium dispensed as:	
Done by :			Checked by:		
*Supplement added before dispensing (if any) -					

Sterilization Details			
Autoclave - ID -		Autoclave Load No.	
Sterilization Cycle Parameter		Prepared / Sterilized Medium Lot Number	
pH of sterilized medium/After Boiling			
Had there been any change in the colour of the sterilized medium			YES / NO
Pink colour formed to 1/3 volume of total volume in Fluid Thioglycollate Medium			YES / NO / NA
Done By		Checked By	

STANDARD OPERATING PROCEDURE

Title: Preparation Storage and Use of Culture Media				
SOP No.:		**Department:**	Microbiology	
		Effective Date:		
Revision No.:	00	**Revision Date:**		
Supersede Revision No.:	Nil	**Page No.:**	16 of 20	

Plates Preparation and Pre incubation Details			
LAF ID -		Incubator ID -	
Supplement added after sterilization (if any) -			

Plates prepared by	No. of plates prepared	Preincubation Details				
		Preincubation Done by (Date /Sign)	Observation Done by (Date /Sign)	Number of Contaminated Plates	No. of plates for usage	Checked by (Date/Sign)

Growth Promotion Test Detail Incubation Conditions - Bacteria - 32.5 ± 2.5º C for 48 - 72 hrs and Fungi - 22.5 ± 2.5º C for 72 - 120 hrs															
Culture Suspension Prepared On - _____		Organism Tested												Negative Control **	
Dilution Used -															
Test (T)	Duplicate (D)	T	D	T	D	T	D	T	D	T	D	T	D	T	D
Observation ***															
Observed done by Date / Sign															
Performed By						Checked By									
RESULT: The Growth Promotion Test for the medium PASSES / FAILS															

** Incubation for 14 days incase of sterility media.

*** '+ ve' Indicates Growth, '-ve' Indicates No Growth.

Remarks - The medium prepared is qualified. YES / NO

Reviewed by
(Sign and Date)

ANNEXURE - V
MEDIA PREPARATION AND USAGE RECORD – II

Date of preparation _____ Use before: _____ Lot No: _____

Media Details:

Name of Media_____ Make: _____ Code No.: _____

Batch No.:_____ Mfg date: _____ Expiry Date: _____

A) Preparation details:

Quantity prepared: _____ml

Weight of dehydrated Media taken : _____gm

Or (if media prepared form basic ingredient)

Name of ingredient:	: Weight	Name of ingredient:	: Weight
_____	:_____gm	_____	:_____gm
_____	:_____ gm	_____	

Balance Used ID. No: _____

Volume of Water for injection adds. : _____ml

pH of media :_____ pH meter used ID No._____

Done by_____ Checked by_____

B) Sterilisation details:

Sterilisation date: _____Sterilisation hold time_____temprature_____

Autoclave Cycle No. : _____Autoclave used ID No._____

Done by_____ Checked by_____

If any delay in distribution after sterilisation: Hold time_____ temperature_____

C) Distribution details:

Laminar air fellow (LAF) used ID No._____

Done by_____ Checked by_____

D) Growth promotion test:

Date of test: _____

Suspension prepared on: _____

Challenge organism	Laboratory code of Suspension used	Colonies added	Replicate	Observation Date				
			I					
			II					
			Obs. By					
			I					
			II					
			Obs. By					

+ = Growth - = no Growth

Incubator used ID No : _____

LAF used ID No : _____

Date of completion : _____

Remarks: Prepared media lot under test is suitable / not suitable for use

Done by Checked by

(Sign & Date) (Sign & Date)

Media consume on_____ or Discontinue due to_____ and Discarded on _____

Tested by Checked by

(Sign & Date) (Sign & Date)

ANNEXURE - VI
CULTURE DILUTION / SUSPENSION LABEL

Culture Dilution / Suspension Label

Name of Organism: _____

MTCC/ATCC No.: _____

Passage No.: _____

Serial Dilution: _____Concentration: _____

Dilution / Suspension Prepared On: _____

Result Obtained On: _____Valid up to: _____

Due date: _____ Sign: _____

ANNEXURE - VII
CULTURE SUSPENSION PREPARATION RECORD

Culture Suspension / Dilution Prepared On: _____ Results Obtained On: _____
Valid Up to: _____ Use Before Date: _____ Due date of Culture suspension preparation: _____

Working Culture Passage No.: _____

Test Organisms	MTCC/ Equivalent ATCC No.	Plate No.	Viable Count In Dilutions										* Test suspension / Dilution
			10^{-1}	10^{-2}	10^{-3}	10^{-4}	10^{-5}	10^{-6}	10^{-7}	10^{-8}	10^{-9}	10^{-10}	
		I st plate											
		II nd plate											
		Average											
		I st plate											
		II nd plate											
		Average											
		I st plate											
		II nd plate											
		Average											
		I st plate											
		II nd plate											
		Average											
		I st plate											
		II nd plate											
		Average											

* Test Suspension / Dilution: Dilution to be used for growth promotion test

Medium Name	Sterilized Medium Lot No.

Tested By:
(Date & Sign)

Checked By:
(Date & Sign)