

CHAIN FREEZING OF RECTAL SWABS AND ISOLATED ORGANISMS.



CHN59: FREEZING OF RECTAL SWABS AND ISOLATED ORGANISMS.

1.0 PURPOSE / INTRODUCTION:

1.1 Introduction:

Patients samples that may need freezing for culture and organisms isolated from culture specimens that are considered significant pathogens are frozen in Sarstedt/Nunc 1.8 ML freezing vial containing Tryptone Soy Broth with 15 % glycerol and stored at -80°C.

1.2 Purpose:

This SOP provides guidance on how to freeze swabs and isolated microorganisms from rectal swabs in the microbiology section.

2.0 SCOPE / RESPONSIBILITY:

This SOP is applicable to all technologists working for the CHAIN study in the Microbiology laboratory.

3.0 SAFETY/RISK ASSESSMENT:

Handle *Salmonella species*, *Shigella species* in the biosafety cabinet.

4.0 DEFINITIONS:

Pathogens: Disease causing organisms.

5.0 SPECIMEN:

Culture plate with a pure growth of the isolated organism.

6.0 EQUIPMENT / MATERIALS/ REAGENTS:

6.1 -80°C freezer.

6.2 Sarstedt vial/Nunc 1.8 ml cryotube with Tryptone Soy Broth and 15% glycerol.

6.3 Bunsen burner.

6.4 Nichrome wire loop.

7.0 METHODOLOGY:

7.1 Stick the specific printed barcode label on the cryotube

7.2 Sterilize the nichrome wire loop by heating, allow to cool and pick enough colonies from culture plate.

7.3 Transfer the colonies into the freezing vial by rubbing the loop onto the sides of the vial to obtain a turbid suspension. In the case of rectal stool swab number 2 (R2), insert the swab sample into the 2ml tube containing freezing mixture cut the swab at the recommended height and cap the tube immediately.

7.4 Transfer the vial immediately and place at the assigned position in a study specific labeled tray in the -80°C freezer.

NB: - Make sure that, the organism being frozen is a pure culture and sufficient enough for freezing.

- For the preparation of Tryptone Soy Broth with 15% glycerol, refer to Appendix 8.1

8.0 APPENDICES:

8.1 Preparation of Tryptone Soy Broth with 15% glycerol (Freezing Mixture)

8.1.1 In a cornical flask, dissolve 3.0g of TSB powder in 85mls of distilled water.

8.1.2 Add 15mls of glycerol and distribute 1ml volumes of the mixture in Sarstedt vials

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arranged in a metal rack.

8.1.3 Recap the vials loosely and sterilize by autoclaving at 121°C for 15 minutes.

8.1.4 Tighten the caps after sterilization and place the vials to cool on the bench in the media preparation room.

8.1.5 Label the rack (freezing mixture), date of preparation and expiry and then place it in the media fridge.

9.0 REFERENCES:

9.1 Practical medical microbiology by Mackie and McCartney 13th Edition.

9.2 Medical laboratory Manual for Tropical countries, volume 11 by Monica Cheesbrough.

Document history

Version	Author	Approved by	Dated
1.01 (MASTER) FREEZING OF RECTAL SWABS AND ISOLATED ORGANISMS.CHN	Joseph Waichungo	Caroline Tigoi	10/10/2016
1.02			

Site training record

All sites are required to maintain a master copy of this SOP that documents the site staff that have been trained on this SOP.

Document History				
Version No.	Trained staff initials	Signature of trained staff	Date	Trainer's Initials
1.01	KDT	Example row	1 st Jan 2016	DM