HEALTH HOLDING

HAFER ALBATIN HEALTH CLUSTER
MATERNITY AND CHILDREN HOSPITAL

Department:	Laboratory and Blood Bank (Microbiology)		
Document:	Internal Policy and Procedure		
Title:	Media Preparation		
Applies To:	All Laboratory Staff		
Preparation Date:	January 05, 2025	**Index No:**	LB-IPP-145
Approval Date:	January 20, 2025	**Version :**	2
Effective Date:	February 20, 2025	**Replacement No.:**	LB-IPP-145(1)
Review Date:	February 20, 2028	**No. of Pages:**	03

1. PURPOSE:

1.1 To establish system and responsibilities for preparation of Media & reagents.

2. DEFINITIONS:

2.1 MSDS: material safety data sheet.

3. POLICY:

3.1 Prepared only in case of shortage in ready-to-use media.
3.2 Use only ingredients that are suitable for microbiology use.
3.3 Do not open new containers of dehydrated media before finishing previous ones.

4. PROCEDURE:

4.1 **Instructions for preparation of media:**

4.1.1 Follow instructions manual provided for preparation of each type of media.
4.1.2 Use only ingredients that are suitable for microbiology use.
4.1.3 Do not open new containers of dehydrated media before finishing previous ones.
4.1.4 Weigh accurately in a clean dry and dust free atmosphere. Once the ingredients are weighed, do not delay in making up the medium.
4.1.5 Use completely clean glassware. The capacity of container should be twice the medium being prepared.
4.1.6 Use distilled water. If distilled water is not available, use deionized water provided exchange resin do not contain substance inhibitory to bacteria.
4.1.7 Add the powder to the water (not water to the powder).
4.1.8 Mix by continues stirring or by rotating the flask.
4.1.9 When heating is required to dissolve, stir while heating (avoid boiling or overheating).
4.1.10 Autoclave a medium only when the ingredients are completely dissolved.
4.1.11 PH of the medium adjusted as directed.

4.2 **Sterilizing culture media:**

4.2.1 Most of the media sterilized by **autoclaving at 121 °C for 15 - 20 min**. This ensures the destruction of bacterial endospores and vegetative cells.
4.2.2 **For XLD agar**: Heat with frequent agitation until the medium boils (do not autoclave or overheat).Transfer immediately to water bath at 50°C.
4.2.3 **For TCBS agar**: Bring to the boil until completely dissolved, do not autoclave, allow to cool to about 50 ^{0}C.
4.2.4 **Dispensing sterile media into Petri-dish:** Dispense media in a clean, dust free room, by using aseptic technique.
4.2.5 Mix the medium gently by rotating the flask. Flame sterilizes the neck of the flask and pour about 15ml of medium in each dish. If air bubbles enter, flame the surface before gelling.
4.2.6 Clearly label the culture media. Store at 2-8 °C when gelled and cooled.
4.2.7 Perform sterility testing before use.

4.3 **Labelling and storage of culture media:**

4.3.1 When the medium has gelled and cooled, stack the plates and label them by the type of medium and the expiration date.

4.3.2 Seal the plates in plastic bags to prevent loss of moisture and reduce the risk of contamination.

4.3.3 Don't leave the plates exposed to bright light especially sunlight.

4.3.4 Plates of culture media should be stored at 2-8 °C in sealed plastic bags.

4.3.5 Most media in screw -cap tubes or bottles can be stored at room temperature (20-24 °C).

4.3.6 Shelf- life:

4.3.6.1 For solid media ----- one month.

4.3.6.2 For liquid media ------ 6 months.

(Providing there is no change in the appearance of medium to suggest contamination or deterioration).

4.4 **Quality control of culture media:**

4.4.1 Sterility testing: Media in Petri dishes are examined by incubating of 2 plates at RT and another 2 plates in incubator for 48 hours before use.

4.4.2 Performance testing: Whenever possible, all culture media should be tested by set of control organisms (Quality control strains) to check the qualification and goodness of these media.

4.5 **Preparation of reagents:**

4.5.1 Label the container clearly with a water-proof marker. Include the full name of the reagent, its concentration, date of preparation& its expiration date. Protect the label by covering it with clear adhesive tape. If a reagent is harmful, irritant, toxic, corrosive, or flammable, indicate this on the container label (according to MSDS).

4.5.2 Error in the preparation of stains or reagents are due to:

4.5.2.1 Incorrect preparation techniques.

4.5.2.2 Calculating incorrectly the weight or volumes of the constituents.

4.5.2.3 Making dilution errors.

4.5.3 Following the preparation of stain or reagent, its performance must be tested before it is put into routine use.

5. MATERIAL AND EQUIPMENT:

5.1 Weighing scale, heater, and glass wares.

5.2 Autoclave

5.3 Sterile Petri dish

5.4 Media & reagents prepared in our microbiology laboratory

6. RESPONSIBILITIES:

6.1 The assigned technician/ technologist for microbiology section.

7. APPENDICES:

7.1 N/A

8. REFERENCES:

8.1 Procedure Manual, Toronto Medical laboratories / Mount Sinai Hospital department of microbiology

8.2 Bailey & Scott's Diagnostic Microbiology. Feingold & Baron;12th. Ed.2007, C.V. Mosby Co. p. 301.

8.3 Clinical Microbiology Procedures Handbook, American Society of Microbiology, Washington DC, 2005

9. APPROVALS:

	Name	Title	Signature	Date
Prepared by:	Dr. Kawther M. Abdou	Consultant & Lab. Medical Director		January 05, 2025
Reviewed by:	Ms. Noora Melfi Alanizi	Laboratory & Blood Bank Director		January 08, 2025
Reviewed by:	Mr. Abdulelah Ayed Al Mutairi	QM&PS Director		January 13, 2025
Reviewed by:	Dr. Tamer Mohamed Naguib	Medical Director		January 14, 2025
Approved by:	Mr. Fahad Hazam Alshammari	Hospital Director		January 20, 2025