HEALTH HOLDING

HAFER ALBATIN HEALTH CLUSTER
MATERNITY AND CHILDREN HOSPITAL

Department:	Laboratory and Blood Bank (Microbiology)		
Document:	Internal Policy and Procedure		
Title:	Antibiotic Susceptibility Testing and Reporting		
Applies To:	All Laboratory Staff		
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1. PURPOSE:

1.1 To establish system and set responsibilities for processing and reporting of Antibiotic Susceptibility Testing.

2. DEFINITONS:

2.1 **Susceptible (S):** The "susceptible" category implies that isolates are inhibited by the usually achievable concentrations of antimicrobial agent when the dosage recommended to treat the site of infection is used.

2.2 **Intermediate (I):** The "intermediate" category includes isolates with antimicrobial agent MICs that approach usually attainable blood and tissue levels, and for which response rates may be lower than for susceptible isolates.

2.3 **Resistant (R):** The "resistant" category implies that isolates are not inhibited by the usually achievable concentrations of the agent with normal dosage schedules, and/or that demonstrate MICs or zone diameters that fall in the range where specific microbial resistance mechanisms (e.g., ß- lactamases) are likely, and clinical efficacy of the agent against the isolate has not been reliably shown in treatment studies.

2.4 **Test/Report Group A**: "As listed in Tables 1A, IB, and IC", agents in Group are considered appropriate for inclusion in a routine, primary testing panel, as well as for routine reporting of results for the specific organism groups.

2.5 **Test/Report Group B** comprises agents that may warrant primary testing. However, they may be reported only selectively, such as when the organism is resistant to agents of the same class, as in Group A. Other indications for reporting the result might include a selected specimen source (e.g., a third-generation cephalosporin for enteric bacilli from cerebrospinal fluid (CSF] or trimethoprim-sulfamethoxazole for urinary tract isolates); a polymicrobial infection; infections involving multiple sites; cases of patient allergy, intolerance, or failure to respond to an agent in Group A.

2.6 **Test/Report Group C** comprises alternative or supplemental antimicrobial agents that may require testing in those institutions that harbour endemic or epidemic strains resistant to several of the primary drugs (especially in the same class, e.g., B-lactams); for treatment of patients allergic to primary drugs; for treatment of unusual organisms (e.g., chloramphenicol for extra intestinal isolates of Salmonella spp.).

2.7 **Test/Report Group U ("urine")** lists certain antimicrobial agents (e.g., nitrofurantoin and certain quinolones) that are used only or primarily for treating urinary tract infections. These agents should not be routinely reported against pathogens recovered from other sites of infection. Other agents with broader indications may be included in Group U for specific urinary pathogens (e.g., P. aeruginosa and ofloxacin).

2.8 **Test/Report Group O ("other")** includes agents that have a clinical indication for the organism group, but are generally not candidates for routine testing and reporting.

2.9 **Test/Report Group Inv.** ("investigational") includes agents that are investigational for the organism group and have not yet been approved by the FDA.

3. POLICY:

3.1 Antibiotics susceptibility testing and reporting according to the Clinical and Laboratory Standards Institute **(CLSI)** standards.

4. PROCEDURE:

4.1 **Medium:**
- 4.1.1 Disk diffusion: Mueller-Hinton agar (MHA), used only for selected organisms.
- 4.1.2 MIC methods: Microscan panels/ Vitek 2 sensitivity cards

4.2 **Inoculum:** Growth method or direct colony suspension, equivalent to a 0.5 McFarland standard

4.3 **Incubation:** at 35+2 °C; ambient air;
- 4.3.1 Disk diffusion: 16 to 18 hours.
- 4.3.2 MIC methods: 16 to 20 hours.

4.4 **Selective Reporting: (yet not applicable on LIS)**
- 4.4.1 Routine reporting: Group (A) agents.
- 4.4.2 Selective reporting: Groups (B) &(C) agents reported selectively in the following conditions:
  - 4.4.2.1 When the organism is resistant to agents of the same class, as in Group A.
  - 4.4.2.2 Specific specimen source (e.g., a third-generation cephalosporin for enteric bacilli from cerebrospinal fluid (CSF] or trimethoprim-sulfamethoxazole for urinary tract isolates);
  - 4.4.2.3 A polymicrobial infection; infections involving multiple sites;
  - 4.4.2.4 In cases of patient allergy.
  - 4.4.2.5 For urine samples Group (U) is reported.
- 4.4.3 Unexpected resistance: when confirmed, should be reported (e.g., resistance to a secondary agent but susceptibility to a primary agent).

4.5 **General Comments:**
- 4.5.1 For disk diffusion
  - 4.5.1.1 Measure the diameter of the zones of complete inhibition (as judged by the unaided eye), including the diameter of the disk. Hold the Petri plate a few Inches above a black, non-reflecting background illuminated with reflected light.
  - 4.5.1.2 The zone margin should be considered the area showing no obvious, visible growth that can be detected with the unaided eye. Ignore faint growth of tiny colonies that can be detected only with a magnifying lens at the edge of the zone of inhibited growth.
  - 4.5.1.3 Strains of Proteus spp. may swarm into areas of inhibited growth around certain antimicrobial agents. With Proteus spp., ignore the thin veil of swarming growth in an otherwise obvious zone of growth inhibition.
  - 4.5.1.4 With trimethoprim and the sulphonamides, antagonists in the medium may allow some slight growth; therefore, disregard slight growth (20% or less of the lawn of growth) and measure the more obvious margin to determine the zone diameter.
- 4.5.2 When **faecal isolates of Salmonella and Shigella spp**. are tested, only ampicillin, a fluoroquinolone, and trimethoprim-sulfamethoxazole should be reported routinely. In addition, for **extra intestinal isolates of Salmonella spp.**, a third-generation cephalosporin should be tested and reported, and chloramphenicol may be tested and reported if requested.
- 4.5.3 Cefotaxime and ceftriaxone should be tested and reported on **isolates from CSF** in place of cefazolin.
- 4.5.4 **Warning: For Enterococcus spp.**, cephalosporin, aminoglycosides (except for high-level resistance screening), clindamycin, and trimethoprim-sulfamethoxazole may appear active in vitro, but are not effective clinically and should not be reported as susceptible.
- 4.5.5 Organisms that are susceptible to tetracycline are also considered susceptible to doxycycline and minocycline.
- 4.5.6 Only results of testing with ampicillin, one of the third-generation cephalosporin; chloramphenicol; and meropenem should be reported routinely with **CSF isolates of H. influenza**.
- 4.5.7 The results of ampicillin susceptibility tests should be used to predict the activity of amoxicillin. The majority of isolates of H. influenza that are resistant to ampicillin and amoxicillin produce a TEM-type B-lactamase.

4.5.8 Rx: Rifampin should not be used alone for antimicrobial therapy.

4.5.9 Penicillin and ampicillin are drugs of choice for treatment of B-haemolytic streptococcal Infections. Susceptibility testing of penicillin and other B-lactams approved by the FDA for treatment of B-hemolytic streptococcal infections need not be performed routinely, because non-susceptible isolates (i.e., penicillin MICs > 0.12 and ampicillin MICs > 0.25 ug/mL) are extremely rare in any B-hemolytic streptococcus and have not been reported for Streptococcus pyogenes. If testing is performed, any B-hemolytic streptococcal isolate found to be non-susceptible should be re-identified, retested, and, if confirmed, submitted to a public health laboratory.

4.5.10 Rx: Recommendations for intra-partum prophylaxis for Group B streptococci are penicillin or ampicillin. Although cefazolin is recommended for penicillin-allergic women at low risk for anaphylaxis, those at high risk for anaphylaxis may receive clindamycin or erythromycin. Group B streptococci are susceptible to ampicillin, penicillin, and cefazolin, but may be resistant to clindamycin and/or erythromycin. When Group B Streptococcus is isolated from a pregnant woman with severe penicillin allergy (high risk for anaphylaxis), clindamycin and erythromycin should be tested and reported.

5. MATERIALS & EQUIPMENTS:

5.1 Routine culture media.
5.2 Antibiotic discs
5.3 Microscan WalkAway combo panels/ Vitek 2 system AST cards
5.4 Suggested Groupings of Antimicrobial Agents with FDA Clinical Indications That Should Be Considered for Routine Testing and Reporting on Non-fastidious Organisms
5.5 Suggested Groupings of Antimicrobial Agents with FDA Clinical Indications That Should Be Considered for Routine Testing and Reporting on Fastidious Organisms.
5.6 Suggested Groupings of Antimicrobial Agents That Should Be Considered for Routine Testing and Reporting on Anaerobic Organisms
5.7 **Zone Diameter and MIC Interpretive Standards for:**
- 5.7.1 2A. Enterobacteriaceae
- 5.7.2 2B-1. Pseudomonas aeruginosa
- 5.7.3 2B-2. Acinetobacter spp.
- 5.7.4 2B-3. Burkholderiacepacia
- 5.7.5 2B-4. Stenotrophomonas maltophilia
- 5.7.6 2B-5. Other Non-Enterobacteriaceae
- 5.7.7 2C. Staphylococcus spp.
- 5.7.8 2D. Enterococcus spp.
- 5.7.9 2E. Hemophilus influenzae and Hemophilus parainfluenzas
- 5.7.10 2F. Neisseria gonorrhoeae
- 5.7.11 2G. Streptococcus pneumonia
- 5.7.12 2H-1. Streptococcus spp. B-Hemolytic Group
- 5.7.13 2H-2. Streptococcus spp. Viridans Group
- 5.7.14 2I. Neisseria meningitides
- 5.7.15 2J. Anaerobes

5.8 Instructions for Use of Tables

6. RESPONSIBILITIES:

6.1 The assigned technician technologist for the microbiology section.
6.2 The C. pathology specialist/ consultant.

7. APPENDICES:

7.1 N/A

8. REFERENCES:

8.1 CLSI document M100-35th ed. Wayne, PA: Clinical and Laboratory Standards Institute; 2025.

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 12, 2025
Reviewed by:	Dr. Tamer Mohamed Naguib	Medical Director		January 13, 2025
Approved by:	Mr. Fahad Hazam Alshammari	Hospital Director		January 20, 2025