

Identification of Multi-Drug Non-Susceptible Gonorrhea in Massachusetts

A Public Health Laboratory Perspective

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Overview

Gonorrhea Introduction

- Disease & Public Health Importance

MA Sexually Transmitted Disease (STD) Program

- Focus on Gonorrhea

Gonorrhea Testing

- MA Clinical Microbiology Laboratory Test Algorithm

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- MA results → Collaborator results
- Interpretation

Emergent Situation

- Response
- Routine surveillance

Partnerships

- Internal/External

Gonorrhea Overview

Sexually Transmitted Infection (STI) caused by bacterium *Neisseria gonorrhoeae* (also referred to as GC or gonococcus)

- Urethra, rectum, throat

Public Health Importance:

- Infected are often asymptomatic = not seeking treatment
- Untreated infection can lead to serious health problems:
 - Increased pregnancy complications
 - Infertility
 - Unknowing transmission to others > accelerated spread of infection > increased risk of eventual treatment failure
 - Disseminated gonococcal infection
- Currently there is *one* recommended treatment option remaining
 - Ceftriaxone 500mg IM

MA STD Program

Prevention Mission

- Detect/respond to STI in MA
- Reduce morbidity
- Address barriers to health using health equity lens

Regulatory Requirements

- Reporting of all GC infections
- Submission of GC isolates to MA SPHL for surveillance testing

Outreach

- Partner with community health centers providing specific care for at-risk groups
- Provide laboratory support to the clinics
- Specific care

Improve Local/State Capacity to Respond to Antimicrobial Resistant GC (ARGC)

GC Testing Overview

Nucleic Acid Amplification Test (NAAT)

- Pros
 - Sensitive and specific
 - Ease of handling samples
 - Fast turn around time (TAT)
- Cons
 - Detects genetic material, does not indicate an active infection
 - Does not yield a viable organism for further testing such as antimicrobial susceptibility testing (AST)

Culture

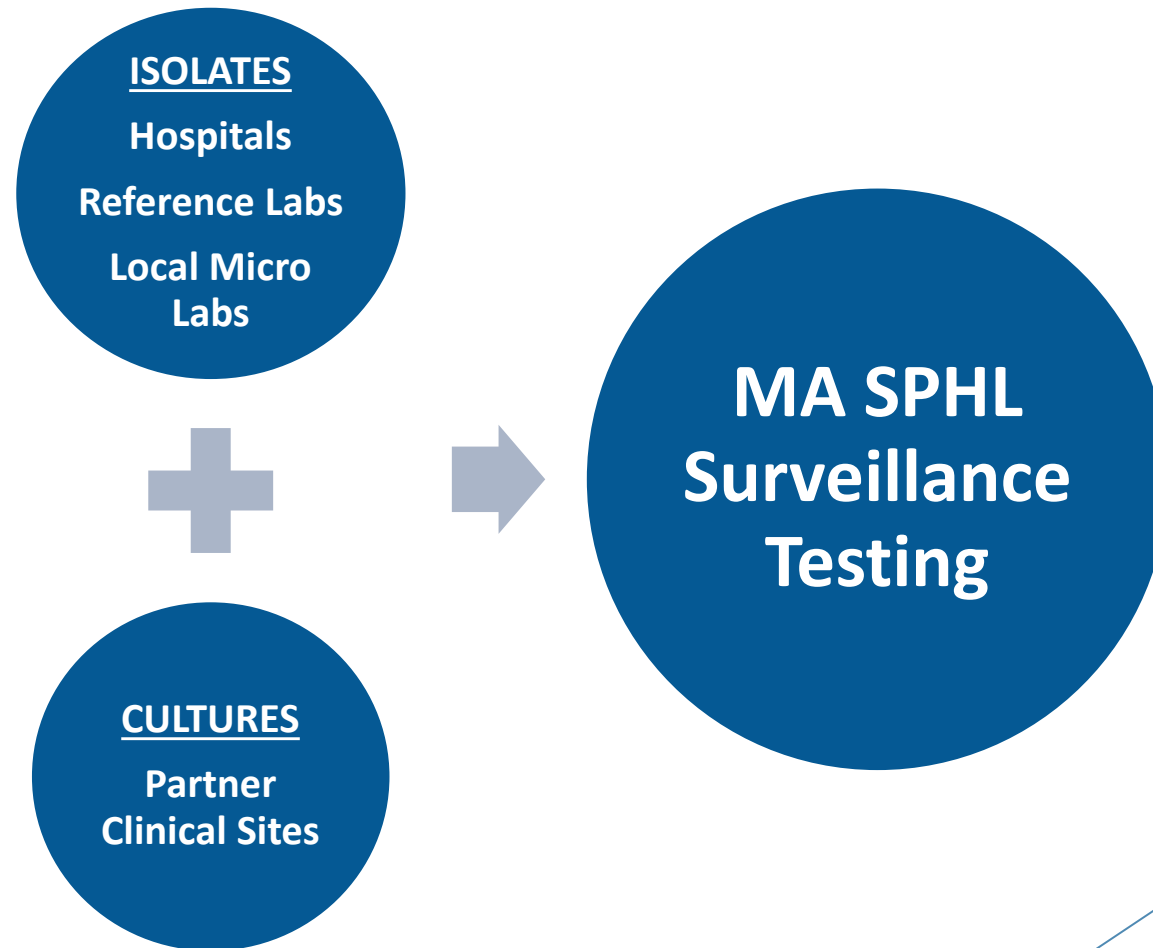
- Pros
 - Positive cultures yield a viable isolate available for downstream testing and characterization
- Cons
 - Transport: sensitive to the cold
 - Special growth conditions: humidity and increased CO₂

MA SPHL Clinical Microbiology Laboratory (CM LAB)

Test Algorithm Overview



Sample Receipt



MA SPHL CM Lab Test Algorithm

Specimen Receipt

- Isolates
- Primary cultures/collections

Culture

- Growth
 - CHOC Agar
 - MTM Agar
 - *Note:* BAP or TSA
 - Not optimal for GC, not used in test protocol
- Environment
 - Humidity
 - CO2

MA SPHL CM Lab Test Algorithm

Identification

- **Bench Tests**
 - **Gram Stain**
 - **Oxidase**
- **Definitive ID**
 - **Primary: Bruker Maldi-ToF**
 - **Mass Spectrometry**
 - **Back-up: Remel RapID NH**
 - **Biochemical**

MA SPHL CM Lab Test Algorithm

AST

- Etest AST method
 - Inoculate Remel GC Agar Base with 0.5 MacFarland dilution of test organism
 - Apply E strip > incubate 20-24 hours > read
 - Generates a quantitative value, a minimum inhibitory concentration (MIC)
- Antimicrobials: bioMerieux Etest strips
 - Ciprofloxacin
 - Ceftriaxone
 - Spectinomycin
 - Azithromycin
 - Cefoxitin
 - Cefixime

MA SPHL CM Lab Test Algorithm

Interpretive Categories and MIC Breakpoints ug/mL (CLIS M100 32nd Ed.)			
<u>Antimicrobial Agent</u>	<u>Susceptible</u>	<u>Intermediate</u>	<u>Resistant</u>
Azithromycin	≤ 1	-- report as Nonsusceptible	
Cefixime	≤ 0.25	-- report as Nonsusceptible	
Cefoxitin	≤ 2	4	≥ 8
Ceftriaxone	≤ 0.25	-- report as Nonsusceptible	
Ciprofloxacin	≤ 0.06	0.12-0.5	≥ 1
Spectinomycin	≤ 32	64	≥ 128

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Specimen Receipt

- **GC isolate: 2 distinct morphologies received for surveillance testing**
- **Original source was urine**
 - **More commonly isolated from urethra, rectum, throat**
- **Isolated on a bi-plate**
 - **GC was growing confluent on the blood agar**

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Culture

- 2 unique morphologies
- Submitted isolates were pure

Identification

- Gram Stain: GNDC
- Oxidase: Positive
- Both identified by Maldi-ToF as *Neisseria gonorrhoeae*

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► AST Etest performed

Antimicrobial Etest Strip	Isolate #1 MIC (ug/mL)	Isolate #2 MIC (ug/mL)
Azithromycin	3	2
Cefixime	1.5*	1.5*
Cefoxitin	2	1.0
Ceftriaxone	0.5*	0.38*
Ciprofloxacin	32	6
Spectinomycin	16	12

*Observation of Cefixime or Ceftriaxone MIC > 0.25 ug/mL is rare and requires notification of Supervisor and repeat testing to confirm.

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► AST Repeat Etest Performed

Antimicrobial Etest Strip	Isolate #1 MIC (ug/mL)	Isolate #1 Repeat MIC (ug/mL)	Isolate #2 MIC (ug/mL)	Isolate #2 Repeat MIC (ug/mL)
Azithromycin	3	2	2	1.5
Cefixime	1.5*	1.5*	1.5*	1.5*
Cefoxitin	2	2	1.0	1.0
Ceftriaxone	0.5*	0.5*	0.38*	0.38*
Ciprofloxacin	32	32	6	8
Spectinomycin	16	12	12	12

*Remember: Observation of Cefixime or Ceftriaxone MIC > 0.25 ug/mL is rare

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► AST Interpretation: *N. gonorrhoeae*

Antimicrobial Etest Strip	Isolate #1 MIC (ug/mL)	Interpretive Category	Isolate #2 MIC (ug/mL)	Interpretive Category
Azithromycin	3	NS	2	NS
Cefixime	1.5*	NS	1.5*	NS
Cefoxitin	2	S	1.0	S
Ceftriaxone	0.5*	NS	0.38*	NS
Ciprofloxacin	32	R	6	R
Spectinomycin	16	S	12	S

► S=Susceptible, NS=Non-susceptible, I=Intermediate, R=Resistant

Remember: “Non-Susceptible” is a category used for isolates where only a susceptible breakpoint is designated because of the absence or rare occurrence of resistant strains.

Collaboration with Centers for Disease Control (CDC)

CDC STD Laboratory and Research Branch

Confirmatory Testing

- **Culture**
- **ID**
- **AST: 2 methods performed**
 - **Etest**
 - **Agar Dilution**
 - **Gold standard in AST protocols**

CDC Confirmatory Testing

ID

- Confirmed as *Neisseria gonorrhoeae* (Maldi-ToF)

AST Etest and Agar Dilution

- Confirmed MA SPHL results
 - Non-susceptible to azithromycin, ceftriaxone and cefixime
 - Resistant to ciprofloxacin
 - Additional antimicrobial testing
 - Also resistant to penicillin and tetracycline

AST Summary

***First Case of Multi-Drug Non-Susceptible GC
Isolated in United States**

	Resistant	Non-susceptible
Azithromycin		NS
Cefixime		NS
Ceftriaxone		NS
Ciprofloxacin	R	
Penicillin	R	
Tetracycline	R	

CDC Downstream Testing

Downstream Research Use Only (RUO) Genetic Analysis

Real-time PCR to Detect AMR

Sanger Sequencing

Whole Genome Sequencing

Analysis identified penA-60.001 allele associated with ceftriaxone NS and also MLST 8123

Asia-Pacific lineage

Monitoring Emerging Pathogen

Expand Surveillance

- Additional enhanced surveillance identified a second isolate
 - Molecular testing of NG-positive remnant diagnostic specimens
 - Analysis supports this organism likely has similar reduced susceptibility to cefixime and ceftriaxone
- Expanded surveillance at health center
 - No additional cases of concern

MA Clinical Alert Sent Out 1/19/2023

- In direct response to identification of 2nd case
- Recommending *culture* on symptomatic patients in addition to routine NAAT testing
- Old school culture will enhance surveillance efforts to give us a better picture of antimicrobial susceptibility

Routine Surveillance

Framework of mandated reporting *and* the additional mandated submission of isolates in MA was critical to identifying this strain

Routine AST on all GC

Supports goal of improving local/state capacity to respond to ARGC

Partners: Importance of Relationships



Internal Partners

Lab/EPI

- Routine outreach
- Outbreak response



Community Partners

Submission of culture/isolates



National Partners

CDC

- Confirmatory downstream testing
- Collaborate on outbreak response

APHL

- Public forum to share knowledge and discuss important public health issues

Takeaways

ARGC is here.

It is our relationships and partnerships that most support response to emerging pathogens.

For PHL teams: your work has real world impact in real time!

Connect with DPH



@MassDPH



Massachusetts Department of Public Health



mass.gov/dph