Challenge M234-2

February 2024

Vaginal/rectal swab (GBS screen): *Streptococcus agalactiae*

HISTORY

A simulated vaginal/rectal swab sample collected from a 35 year old 35 week pregnant woman was sent to category A laboratories. Participants were expected to isolate and report *Streptococcus agalactiae.*

CMPT QA/QC/STATISTICS

All simulated vaginal/rectal samples are produced at CMPT according to CMPT internal protocols. The sample contained a pure culture of *Streptococcus agalactiae.*

The samples are assessed for homogeneity and stability using in-house quality control methods and random selection of samples before and during production, and post sample delivery. The number of random samples selected is 15% of the total production batch.

The challenge sample lot was confirmed to be homogeneous and stable for 18 days.

Organism identification was confirmed by a reference laboratory.

All challenge components have in-house assigned values based on the most clinically appropriate result; the most clinically appropriate result is determined by expert committee evaluation. No further statistical analysis is performed on the results beyond that described under "Suitability for grading."

SURVEY RESULTS

Reference laboratories: 12/12 (100%) labs reported Group B Streptococcus (*S. agalactiae*), 1 reference lab indicated it would refer the sample for identification.

Participants: 49/49 (100%) reporting labs reported Group B Streptococcus (*S. agalactiae*),

Suitability for Grading

A challenge is considered suitable for grading if agreement is reached by 80 percent of selected reference group and at least 50 percent of the participants.

Organism identification was correctly performed by at least 80 percent of reference laboratories and greater than 50 percent of all laboratories. Thus the challenge was determined to be suitable for grading.

MAIN EDUCATIONAL POINTS from M234-2

1. Maternal colonization with *Streptococcus agalactiae* or Group B Streptococcus (GBS) in the vagina or rectum of pregnant women is the primary risk factor for early-onset GBS neonatal disease. Universal antepartum screening permits timely appropriate antibiotic prophylaxis.
2. The most recent updated guidelines (March 2020 rev. July 2021) recommend antepartum screening for GBS at 36 0/7 to 37 6/7 weeks of gestation (1), a change from the previous recommendation to screen at 35-37 weeks of gestation.
3. Antimicrobial susceptibility testing (AST) is recommended on all GBS isolates from pregnant women with severe penicillin allergy with Clindamycin as the recommended antibiotic choice for these women.

COMMENTS ON RESULTS

Five labs (1 reference lab and 4 participant labs) indicated they would refer out this specimen for identification. It is unclear why these labs were not able to perform the necessary enrichment, culture and identification steps as GBS Screening is part of most routine Microbiology test menus.

All 49 responding laboratories correctly identified Group B Streptococcus *(S. agalactiae*). Pure growth of characteristic beta-hemolytic colonies on blood agar plates and/or pigmented colonies on chromogenic media were isolated.

Grading

Maximum grade: 4

Reporting the presence of group B Streptococcus was graded 4.

Table 1. Identification results

Reported	Total	Grade
Group B Streptococcus (*S. agalactiae*) ± beta hemolytic	47	4
Streptocoque bêta-hémolytique du groupe B	2	4
refer, snnp	5	ungraded
Total	**54**	

A variety of identification methods were utilized including, gram stain, biochemical testing (CAMP, PYR), growth in selective & differential media (eg. Carrot broth), latex serology and/or MALDI -TOF mass spectroscopy. Only 1 lab reported the use of Nucleic Acid Amplification Testing (NAAT) as a secondary test on colonies isolated by culture.

ISOLATION AND IDENTIFICATION

In March 2020, the American Society for Microbiology (ASM) published a new guideline—Interim Guideline for the Detection and Identification of Group B Streptococcus (GBS) [1] —which replaces the 2010 guidelines published by CDC.

Specimen Collection & Laboratory Testing Recommendations by the Interim Guideline for the Detection and Identification of Group B Streptococcus (2020)

1. Use a **single swab** to obtain a screening specimen first from the lower vagina and then from the rectum without use of a speculum.
2. Collect vaginal-rectal specimens using a flocked swab and place in a liquid-based transport medium such as Amies transport media.
3. Incubate GBS screening specimens in **selective enrichment broth** prior to agar media plating and also prior to performing NAAT. Intrapartum NAAT without enrichment has an unacceptably high false negative rate, ranging from 6.3% to 22%).[2–4]
4. Acceptable enrichment broths include Todd-Hewitt broth, supplemented with selective agents and carrot broth and liquid biphasic Granada medium, which are also selective thereby allowing the GBS enrichment and detection in a single step.[5–7]
5. Culture media and GBS isolation methods should detect both hemolytic and nonhemolytic strains.
6. Report GBS in any quantity from urine cultures from pregnant women during all trimesters.
7. Acceptable phenotypic and proteomic methods of identification of candidate isolates include CAMP test, latex agglutination, and (MALDI-TOF).
8. Latex agglutination directly from enrichment broth and direct-from-specimen immunoassays are unacceptable methods for GBS detection. Although reported to have high specificity (>99%), it has an unacceptable variable sensitivity (65-99%).[8,9]
9. Perform antimicrobial susceptibility testing on all GBS isolates from pregnant women with severe penicillin allergy.
10. Nucleic acid amplification-based identification of GBS from enrichment broth is acceptable, but not sufficient for all patients. NAAT without enrichment has an unacceptably high false negative rate, ranging from 6.3% to 22% (2-4). Hence direct specimen NAAT is not recommended for GBS screening for the purposes of determining intrapartum prophylaxis. Studies are in progress to evaluate when, how, and if a direct-from-specimen NAAT should be used.
11. Since NAAT detection of GBS does not produce an isolate for AST testing, the method is **not** sufficient for pregnant women who have penicillin allergy and at risk for severe anaphylaxis, as they cannot receive penicillin or cefazolin for intrapartum prophylaxis.

Important Alert: A note of caution has been raised with the use of molecular diagnostic methods, as mutants that have lost target nucleic acid sequences have been detected. A mutant with deletion in the CAMP factor (*cfb*) gene is of particular concern as 8 of the 10 different nucleic acid amplification tests cleared by the U.S. Food and Drug Administration target the amplification of the *cfb* gene.[10]

Although these diagnostic-escape mutants represented only 1% to 7% of the total GBS isolates in the four geographic locations analyzed,[11,12] more worldwide surveys are needed to truly understand the impact of chromosomal deletions in molecular- evasion mutants.[13]

ANTIMICROBIAL SUSCEPTIBILITY

Antimicrobial susceptibility testing (AST) is recommended on all GBS isolates from pregnant women with severe penicillin allergy with Clindamycin as the recommended antibiotic choice for these women. CLSI recommends AST for erythromycin and clindamycin given the rising rates of resistance. Erythromycin AST is used to screen for possible inducible clindamycin resistance; isolates demonstrating erythromycin resistance and clindamycin-susceptible or clindamycin-intermediate results are further tested for inducible clindamycin resistance. The AST report must include results for clindamycin (including inducible clindamycin resistance) and vancomycin.

Reporting of vancomycin may be considered if the isolate is not clindamycin susceptible and if the patient is penicillin allergic. If susceptibility testing can't be performed on all isolates from pregnant women, the guidelines recommend that electronic screening culture orders include "indication of penicillin allergy" as a mandatory field.

Since intrapartum GBS prophylaxis consists of systemic antimicrobials, the choice of agents used is different from those used to treat urinary tract infection with Group B Streptococcus. Urine culture isolates from penicillin-allergic pregnant women should, similarly, be tested for clindamycin resistance, with the understanding that clindamycin is inappropriate for treatment of a urinary tract infection, but, rather, could be used as intrapartum prophylaxis

CLINICAL RELEVANCE

Streptococcus agalactiae or Group B Streptococcus (GBS) has long been recognized as the primary cause for neonatal infection, defined as early-onset disease (EOD) with case-fatality rates as high as 50%. Maternal colonization with GBS in the vagina or rectum of pregnant women is the primary risk factor for transmission during delivery and infection of the neonate.

There are two clinical manifestations of invasive GBS disease in the neonate, classified as early-onset (EOD) and late-onset disease. EOD comprises approximately 60% of infections,[14] presents within 7 days of birth, and occurs secondary to vertical transmission during labor and delivery. It usually manifests within the first 12 to 48 hours after delivery, commonly presenting with sepsis and pneumonia, and less commonly with meningitis; 20% of infants surviving GBS meningitis are left with moderate to severe neurodevelopmental impairment.[15]

Late-onset GBS disease presents between >7 and <90 days of age, 7 and occurs secondary to horizontal transmission from the mother, hospital sources, or individuals in the community. [14]

Maternal intrapartum GBS colonization is the primary risk factor for early-onset (i.e. onset <7 days of life) of invasive group B streptococcal disease in infants. In the absence of any intervention, an estimated 1%-2% of infants born to colonized mothers develop early-onset GBS infections.[16] Approximately 10-30% of pregnant women are colonized with GBS in the vagina or rectum.[16]

Other factors that increase the risk for early-onset disease include gestational age <37 completed weeks, longer duration of membrane rupture, intra-amniotic infection, young maternal age, black race, and low maternal levels of GBS-specific anticapsular antibody.[17,18]

The most recent updated guidelines (March 2020 rev. July 2021) recommend antepartum screening for GBS at 36 0/7 to 37 6/7 weeks of gestation (1), a change from the previous recommendation to screen at 35-37 weeks of gestation. The predictive value of prenatal cultures for GBS detection decreases significantly when collected more than 5 weeks before delivery. Universal screening 5 weeks prior to delivery (at 36-37 weeks gestation) for maternal GBS colonization and use of intrapartum antibiotic prophylaxis has resulted in substantial reductions in early-onset GBS disease among newborns.[17]

While the object of screening is to enable the prevention of neonatal infection, pregnant women are at increased risk of GBS infection, and the incidence is about double that of non-pregnant women. Most maternal infections occur during labor and delivery, but if earlier, infection may result in premature labor and sepsis in neonates.[19] [15]

GBS is an important pathogen in the elderly, especially when other comorbidities are present, but a full description of GBS infection in this group is beyond the scope of this critique.

History of guidelines: 1996, 2002, 2010, 2019/20 has been revised in previous critiques (M212-2)

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