AMERICAN SOCIETY FOR MICROBIOLOGY | Journal of Clinical Microbiology®

MINIREVIEW

Reliable Detection of Group B Streptococcus in the Clinical Laboratory

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ABSTRACT Group B streptococcus (GBS) is a leading cause of invasive neonatal infections and a significant pathogen in immunocompromised adults. Screening to detect GBS colonization in pregnant women determines the need for antibiotic prophylaxis in that pregnancy. Efficient determination of the GBS colonization status of pregnant women is crucial. Methods that maximize the probability of GBS recovery are needed. The availability of technologies such as matrix-assisted laser desorption ionization–time of flight mass spectrometry (MALDI-TOF MS), molecular techniques, and chromogenic culture media, including Granada-type media, have changed the scenario for GBS detection and identification. This review presents and evaluates novel diagnostic tools, as well as classic identification techniques, for GBS species determination.

KEYWORDS GBS, group B streptococcus

Accepted manuscript posted online 28 June 2017

Citation Rosa-Fraile M, Spellerberg B. 2017. Reliable detection of group B streptococcus in the clinical laboratory. J Clin Microbiol 55:2590–2598. https://doi.org/10.1128/JCM.00582-17.

Editor Colleen Suzanne Kraft, Emory University

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Streptococcus agalactiae, group B streptococcus (GBS), is a Gram-positive encapsulated bacterium that belongs to the group of pyogenic streptococci. It is the only *Streptococcus* species harboring the Lancefield group B cell-wall-specific polysaccharide antigen that is common to all GBS strains. GBS can be subdivided into 10 different serotypes (Ia, Ib, and II to IX) on the basis of type-specific capsular polysaccharides (1).

In most cases, GBS is an asymptomatic colonizer of the digestive and genitourinary tracts of healthy human adults. However, it can cause severe invasive infections in neonates and immunocompromised adult patients. The first reports about GBS as a human pathogen were published in the late 1930s, when three fatal cases of puerperal sepsis caused by GBS were described (2). Since the 1960s (3), GBS has remained a leading cause of life-threatening neonatal infections (1). In neonatology, there are two distinguishable clinical syndromes; early-onset disease (EOD) is a GBS infection occurring within the first week of life (usually within the first 24 h), and late-onset disease (LOD) is a GBS infection presenting after 7 days of age (7 to 90 days postpartum). EOD is caused by vertical transmission of GBS from a colonized mother to her newborn, through either ascending infection from the genital tract or GBS transmission to the newborn during labor and birth. Numerous studies have shown that up to 30% of pregnant women worldwide are colonized with GBS, and vertical transmission occurs for roughly 50% of colonized mothers. About 1% of colonized newborns develop EOD. EOD occurs mainly after the onset of labor or in connection with ruptured membranes, although infection of the fetus can happen through intact membranes. Bacteremia without a focus is the most common clinical syndrome, followed by pneumonia and meningitis. Even today the case fatality rate for EOD is estimated to be 2 to 10%, and fatal outcomes are more frequent among premature neonates (1).

Because most EOD is acquired through contact of the neonate with GBS during delivery, intrapartum antibiotic prophylaxis (IAP) administered to GBS carriers prevents vertical transmission in the vast majority of cases, and its widespread use has resulted in significant reductions in the incidence of EOD (4). In contrast to EOD, LOD is most

likely acquired after birth, from breast milk or from nosocomial or community sources. Prematurity is the main risk factor for developing LOD, and bacteremia without a focus of infection is the most common presentation. The mortality rate for LOD is lower, but meningitis and subsequent sequelae are more frequently associated with LOD (1).

GBS also causes significant maternal morbidity, including endometritis, chorioamnionitis, bacteremia, and postpartum wound infections. GBS urinary tract infections are associated with miscarriages, preterm births, and low-birth-weight newborns (1). Although GBS seldom causes disease in healthy adults, it is responsible for serious infections in diabetics, elderly individuals, residents in nursing homes, and otherwise immunocompromised patients (5). The successful administration of IAP and the treatment of severe GBS infections rely on efficient and reliable detection of GBS in clinical samples. GBS is also important in veterinary medicine, being a main cause of bovine mastitis and a major pathogen in farmed fish (6). Identification of GBS may be accomplished with a variety of microbiological techniques, and Table 1 provides an overview of them and their characteristics.

ANTENATAL DETECTION OF *STREPTOCOCCUS AGALACTIAE*

The first reports showing that administering IAP to mothers who are colonized with GBS (and who are typically asymptomatic) reduces vertical transmission of GBS and prevents GBS EOD were published in the 1980s (7). These findings led to a general CDC recommendation for IAP in 1996, which was rapidly accepted and resulted in a significant decline in EOD cases. A key prerequisite for efficient application of IAP is the reliable detection of GBS vaginorectal colonization before delivery (1–4). Because GBS colonization status can fluctuate during pregnancy, the timing of specimen collection is important. Positive predictive values decrease when a culture is performed more than 5 weeks before delivery. Swabbing both the distal vagina (vaginal introitus) and the rectum increases GBS recovery significantly, compared with swabbing the vagina only. Therefore, to predict GBS colonization status at the time of delivery accurately, the current CDC guidelines recommend screening for GBS by collecting lower vaginal and rectal swabs at 35 to 37 weeks of pregnancy (4).

Swabs should be placed in an appropriate transport medium, such as Amies or Stuart medium, and can be kept in the transport medium for up to 4 days (4). If processing is delayed, refrigeration is highly preferable to storage at ambient temperature, as GBS viability decreases significantly over time (8, 9). The use of flocked swabs has been introduced for collection of bacteriological samples. These swabs incorporate liquid Amies transporting medium and are designed to minimize the entrapment of specimens. Flocked swabs (ESwab collection device; Copan Diagnostics, Brescia, Italy) have been reported to significantly increase the recovery of GBS, compared with the use of standard fiber swabs (9). Swabs should be inoculated into a selective enrichment broth (e.g., Todd-Hewitt broth with gentamicin, nalidixic acid, and sheep blood [Baker broth] or with colistin and nalidixic acid [Lim broth]) and incubated at 35°C to 37°C for 18 to 24 h (4). Selective GBS broths are also commercially available. However, competitive growth studies have found that *Enterococcus faecalis* present in vaginorectal samples can suppress the growth of GBS in selective broths, causing the subculture to test negative for GBS (10, 11). This type of false-negative result can be avoided by the inoculation of an adequate plating medium in addition to the selective enrichment broth. Prior to selective broth inoculation, swabs can be streaked on blood agar, selective blood agar (such as neomycin-nalidixic acid agar [NNA] or colistin-nalidixic acid agar [CNA]), Granada agar, or chromogenic agar. These plates should be incubated at 35°C to 37°C (Granada plates in anaerobiosis) and examined for GBS-like colonies after 24 and 48 h. If GBS colonies are detected, then the selective broth can be discarded, decreasing the time to obtain results (4). After overnight incubation, the selective broth should be subcultured on blood agar, chromogenic agar, or Granada agar and incubated for 24 to 48 h. For chromogenic agar, colored colonies indicative of GBS should be monitored; for Granada agar, the development of brick, orange, or red colonies should be observed. GBS-like colonies that develop in chromogenic agar

TABLE 1 Overview and characteristics of laboratory methods for identification of group B streptococcus

Laboratory method	Special equipment requirements	Sample type	Time to results	Relative sensitivity[a]	Relative specificity[a]	Relative costs[b]	Advantages	Disadvantages	Comments
Beta-hemolysis	None	Clinical samples; isolated colonies required	Overnight incubation to 2 days	++	++	$	Traditional, inexpensive, very easy to perform	Failure to detect nonhemolytic GBS strains	Beta-hemolysis may be difficult to observe
Granada-type media	None	Clinical samples; isolated colonies in Granada agar required	Overnight incubation	++++	+++++	$	Very easy to observe, 100% specific for GBS	Anaerobic incubation, failure to detect nonhemolytic GBS strains	Highly discriminatory; anaerobiosis not required for Granada broths; frequently negative results for nonhuman GBS strains
Chromogenic media	None	Clinical samples; isolated colonies required	1–2 days	++++	++	$	Anaerobic incubation not necessary	GBS-like colonies require confirmatory tests	Sensitivity, specificity, and colony aspects vary among suppliers
CAMP test	None	Isolated colonies; previous culture isolation required	Overnight incubation	++++	+++	$	Very easy to perform, easy to read	Requires 1 additional day to confirm GBS	Also positive for *Streptococcus porcinus* and some group A streptococci
PYR test	None	Isolated colonies	Minutes	Always negative for GBS	Always negative for GBS	$	Easy to perform, commercially available from many suppliers		Always positive for group A streptococci; used to differentiate GBS from group A streptococci, enterococci, and *S. porcinus*
Hippurate	None	Isolated colonies	Minutes	++	+	$	Easy to perform	Too unspecific for identification or confirmation of GBS	Positive results for most GBS but also other streptococci
Biochemical profiling	Manual kits or automated systems	Isolated colonies	Minutes to overnight	+++	++	Test, $$; platform, $$$	Commercially available from many suppliers	Costs	Not widely used for identification of GBS; better to reserve kits for identification of other streptococcal species
MALDI-TOF MS	MALDI-TOF MS instrument	Isolated colonies	Minutes	++++	++++	Test, $; instrument, $$$$	Easy and simple to perform	Requires specialized equipment, with high initial investment costs	Displacing phenotypic identification techniques in laboratories
Latex agglutination/ coagglutination	None	Isolated colonies	Minutes	++++	+++	$	Easy and simple to perform, available from many manufacturers	*S. pseudoporcinus* and atypical enterococci (hemolytic) may cross-react with GBS grouping antisera	
Direct antigen detection	None	Clinical samples	Minutes	+	++	$	Easy to perform, point-of-care test	Sensitivity insufficient for direct detection of GBS in clinical samples	Can help in diagnosis of neonatal GBS meningitis
PCR, clinical sample	Automated PCR platform	Clinical samples	hours	+++	++++	Test, $$; platform, $$$	Can be performed at delivery	No antibiotic susceptibility data	Not CDC recommended for GBS screening purposes
PCR, enrichment broth	Automated PCR platform	Enrichment broth	Hours to overnight	++++	++++	Test, $$; platform, $$$		No clear advantage over culture-based methods	Complies with CDC recommendation
DNA sequencing	Diagnostic molecular biology	Isolated colonies	Days	+++++	+++++	$$$	Gold standard for taxonomic purposes	Requires molecular biology expertise	Very rarely required to identify GBS

[a]Values for sensitivity and specificity estimates may be misleading because they depend on the test used for comparison. Thus, relative sensitivity and specificity are shown (from + to +++++).
[b]Costs can vary according to the location and laboratory testing volume.

should always be confirmed as GBS using additional tests (e.g., the latex agglutination test or the CAMP test) to avoid false-positive results. Typical GBS colonies that are not pigmented on Granada agar should be further tested to rule out nonhemolytic GBS. The latex agglutination test or the CAMP test can be appropriate for this purpose. Subcultures on blood agar plates should be incubated for 18 to 24 h but should be reincubated overnight if typical hemolytic colonies are not observed. GBS beta-hemolysis can be difficult to observe, so typical colonies without hemolysis on blood agar plates should be tested further. Considerable effort is thus required to detect nonhemolytic nonpigmented GBS isolates (4, 12). However, the relevance of nonhemolytic nonpigmented colonizing GBS strains as a source of invasive bloodstream infections is currently under discussion. Very few studies on the role of these strains as a cause of EOD have been published (13, 14). Furthermore, nonhemolytic invasive GBS strains may even originate from beta-hemolytic colonizing strains and arise during bloodstream infections through spontaneous mutations (15).

As an alternative to selective broth, swabs can be inoculated into tubes of Granada-type broth, such as Strep B Carrot broth (Hardy Diagnostics), which is used as an enrichment and detection broth for GBS (9, 16). The detection of red-orange pigment (color change and/or pink or red spots) after 18 to 24 h of incubation is specific for the presence of beta-hemolytic GBS, avoiding subcultures and reducing the detection and identification of GBS to a single step. However, Granada-type broths detect only hemolytic strains. Therefore, negative tubes (no color) should be further tested by either subculture on sheep blood agar plates, direct latex agglutination testing, or use of a nucleic acid amplification technique (NAAT) to detect nonpigmented nonhemolytic GBS strains (4). It has also been proposed to detect GBS directly from incubated Lim or Baker selective broth by using agglutination techniques or molecular tests (4, 17), thus avoiding an additional subculturing step. However, the presence of blood or heavy growth of *E. faecalis*, which may sometimes develop in selective broth, can interfere with GBS detection (11, 18). An algorithm for the CDC-recommended laboratory testing for prenatal screening for GBS colonization is depicted in Fig. 7 of reference 4. Furthermore, it must be noted that culture methods are always required for penicillin-allergic women, because susceptibility testing should be carried out on their antenatal GBS isolates (4).

PHENOTYPIC METHODS FOR IDENTIFICATION OF GBS

Most human GBS isolates grow readily on blood agar after overnight incubation, as large colonies (3 to 4 mm in diameter) with a narrow zone of beta-hemolysis (19). GBS beta-hemolysis may be difficult to detect for some strains, however, and can be observed only when colonies are detached from the blood agar. In most laboratories, colonies displaying typical GBS morphology are subjected to latex agglutination tests to determine the presence of the Lancefield group B antigen and, if they test positive, they are designated GBS. A decision that is based on detection of the Lancefield group B antigen is very specific, since *S. agalactiae* is the only streptococcal species harboring this antigen. However, *Streptococcus porcinus*, which may be present in the genital tract of pregnant women, grows on blood agar as beta-hemolytic colonies and cross-reacts with GBS agglutination kits. For this reason, the detection of beta-hemolytic colonies that are positive in GBS latex agglutination tests requires further testing (20, 21). The zone of beta-hemolysis surrounding the *S. porcinus* colonies on sheep blood agar is usually wider and much more pronounced than that observed with *S. agalactiae* (22, 23). A simple and specific method to distinguish GBS from other beta-hemolytic streptococcal species is detection of the reddish polyenic pigment granadaene. The expression of granadaene is invariably linked to the expression of GBS β-hemolysin because they are encoded by a single genetic locus known as the *cyl* operon (24). Moreover, it has been reported that GBS pigment and β-hemolysin could be the same molecule (25). As no other streptococcal species produce granadaene (6), detection of the pigment can be used as a simple and fully specific method for single-step identification of GBS (26), eliminating the need for ancillary tests. Detection of GBS

pigment is generally carried out on Granada-type media, on which beta-hemolytic strains of GBS produce pigmented colonies. A folate pathway inhibitor (methotrexate) and Bacto Proteose Peptone N3 (BD) in the presence of starch trigger the production of pigment in these media (6). There are two classes of Granada-type culture media. The first is Granada agar, marketed in the United States by Hardy Diagnostics and in the European Union by bioMérieux and BD. The second is Granada broth, marketed by bioMérieux (ChromID Granada Biphasic) and in the United States by Hardy Diagnostics (Strep B Carrot broth). For optimal production of pigment, it is necessary to incubate Granada agar plates anaerobically or with a coverslip placed over the inoculum (27). However, anaerobic incubation is not necessary when using Granada broth. Although Granada-type media incorporate selective agents (crystal violet, colistin, and metronidazole), they are not completely selective for GBS. Other microorganisms, mainly enterococci and yeast, can also grow on the medium and appear as white colonies (27). The identification of GBS strains by detection of hemolysis and pigment production is not 100% sensitive, however, because 2 to 5% of human-colonizing GBS isolates are nonhemolytic and do not produce pigment (4, 12, 28, 29). For such GBS strains, which appear white on Granada-type media, identification has to rely on other tests. Because β-hemolysin represents an important GBS virulence factor, strains lacking pigment and hemolysin production may not have full pathogenic potential (13, 25, 30). Since nonhemolytic and nonpigmented GBS strains are more frequently encountered among animal strains, Granada-type media are unreliable for the detection of GBS in veterinary clinical samples (6).

CHROMOGENIC MEDIA

In recent years, there has been rapid expansion in the availability of chromogenic media for the detection of pathogenic bacteria and yeasts. These culture media contain enzyme substrates linked to indoxyl chromogens, and the target microorganisms are characterized by specific enzyme systems that metabolize the substrate, resulting in release of the chromogen. Subsequently, the indigoid dye formed upon oxidation and dimerization of indoxyl molecules in the presence of oxygen precipitates within the colonies, leading to typical brightly contrasting colors (31). Research over the past few decades has resulted in the release of several commercially available chromogenic media for GBS detection, including media developed by bioMérieux (ChromID Strepto B), Bio-Rad (StrepB Select), Oxoid (Brilliance GBS), and CHROMagar (CHROMagar StrepB). The manufacturers of these media do not disclose the constituents, however, and to the best of our knowledge there is no chromogenic substrate specific for GBS detection. Therefore, these media have to rely on a mixture of chromogens, trying to ensure that other bacteria develop colonies that do not resemble GBS (32–34). These media should be incubated in a normal atmosphere, because anaerobic incubation suppresses the development of colored colonies by hampering the oxidation of the indoxyl compounds. Owing to the chromogenic substrates present, minimal exposure to light during storage is mandatory, and incubation must be carried out in the dark for optimal performance; otherwise, proper color development of isolates on chromogenic agar is unreliable. These media also incorporate undisclosed selective agents in an attempt to increase the selectivity for GBS. Chromogenic media provide a fast and easy approach for GBS identification but are neither 100% specific nor 100% sensitive. The lack of any GBS-specific chromogen limits their specificity, resulting in potential misidentifications. Bacterial species present in rectovaginal samples, such as *Enterococcus* spp., *Streptococcus bovis*, *S. porcinus*, *Streptococcus pseudoporcinus*, *Streptococcus salivarius*, *Streptococcus thoraltensis*, *Streptococcus anginosus*, *Streptococcus pyogenes*, and *Staphylococcus* spp., can develop GBS-like colonies (20, 29, 34–37). Therefore, to avoid false-positive results, the use of GBS chromogenic media requires confirmation of suspected GBS colonies by using additional tests (such as the CAMP test, the latex agglutination test, molecular techniques, or positivity on Granada media).

While chromogenic media do not rely on detection of the GBS pigment and thus are able to detect nonhemolytic GBS strains, they may display other sensitivity problems.

Especially in samples with heavy growth of commensal flora, it can be difficult to detect a small number of GBS colonies (37). Although rare, false-negative results have been reported for GBS strains that form colonies of atypical appearance (36). Of note, confirmative identification of GBS-like colonies on chromogenic media using matrix-assisted laser desorption ionization–time of flight mass spectrometry (MALDI-TOF MS) can sometimes be difficult due to the presence of substances interfering with mass peak signals (37).

CAMP TEST

Almost all clinical GBS isolates (38) produce another cytolytic toxin, the CAMP (Christie, Atkins, Munch-Petersen) factor. This factor is not hemolytic *per se*, although it lyses sheep erythrocytes pretreated with staphylococcal β-lysin (sphingomyelinase). This cytolysin is distinct from the GBS β-hemolysin and pigment. The CAMP test involves streaking the strain to be tested perpendicular to a streak of a strain of *Staphylococcus aureus* on sheep blood agar. A positive reaction appears as a characteristic arrowhead zone of hemolysis adjacent to the place where the two lines come into proximity (39). However, this test is not 100% specific. Many *S. porcinus* strains, which are sometimes isolated from the genitourinary tracts of female patients (23), and some group A streptococci (GAS) can also produce positive CAMP test reactions (40, 41). The fact that the CAMP factor is present in nonhemolytic nonpigmented GBS strains can be used to generate beta-hemolysis in otherwise nonhemolytic strains, by incorporating the staphylococcal β-lysin into blood agar plates. This principle, which is used in GBS Detect (Hardy Diagnostics), facilitates the detection of nonhemolytic GBS on blood agar. The *cfb* gene that encodes the CAMP factor is present in the vast majority of GBS isolates and is exploited for the molecular identification of GBS (42).

PYR TEST

The pyrrolidonyl arylamidase (PYR) test is used for the detection of pyrrolidonyl arylamidase and is available from many different suppliers. This test invariably yields negative results for GBS and can be used to distinguish GBS from beta-hemolytic enterococci, GAS presenting with atypical morphology, and *S. porcinus*, which are PYR positive (22, 23, 41, 43).

HIPPURATE

Most GBS strains hydrolyze sodium hippurate, producing glycine. However, other streptococci, particularly enterococci, also give positive reactions (39, 44). Although the hippurate test is rapid, is easy to perform, and also is available from different suppliers, its lack of specificity precludes its use to reliably identify GBS.

BIOCHEMICAL PROFILING

For many years, established phenotypic tests have remained cornerstones for the identification of bacterial microorganisms in clinical microbiology. Various test systems that integrate multiple conventional phenotypic tests into a single-step procedure have been developed. After the interpretation of results, the biochemical reactions included in the kit system generate a biotype number that is matched against the profile indexes from a database to identify the bacterial species. Examples of biochemical profiling kits for the identification of streptococci are the API Rapid Strep identification system (bioMérieux) and the RapID STR system (Remel). However, the ability of these systems to identify microorganisms depends on the accuracy of their databases of profile indexes and the inclusion of all relevant microbial species. Although these systems can be used to identify beta-hemolytic streptococci, they are not 100% specific. While the accuracy to identify GBS is high, more reliable results may be achieved by using a combination of simpler phenotypic methods, reserving the use of these kits for the identification of other streptococcal species (39).

MALDI-TOF MS

MALDI-TOF MS is an emerging tool for microbial identification. It is based on the analysis of protein profiles generated using nonfragmenting or "soft ionization" tech-

niques, allowing the analysis of large macromolecules characteristic of each microorganism. The accrued spectra, which are protein fingerprints (typically between 2 and 20 kDa), are compared with a database of known spectra. As with any automated system, the accuracy of identification is dependent on the quality and comprehensiveness of the database. The main advantages of this technology are speed and accuracy (21). However, MALDI-TOF MS is restricted today mostly to the identification of pure colonies grown on a solid medium, because generation of a satisfactory mass spectrum requires that an adequate amount of sample be deposited onto a target for analysis. The need to study pure cultures of microorganisms originates from the inability of current software programs to analyze the spectra acquired from mixed cultures (45). Because of that, MALDI-TOF MS is not yet able to identify microorganisms directly from most clinical specimens, such as swabs and wound specimens. This precludes its use for the direct identification of GBS in vaginal and rectal swabs. Today, MALDI-TOF MS has been adopted as a rapid and robust method for accurate microbial identification in many clinical microbiology laboratories, and it is beginning to supersede phenotypic identification techniques (46). In regard to beta-hemolytic streptococci, including GBS, MALDI-TOF MS-based identification is fast (minutes) and 100% accurate (47). However, while few laboratory consumables are required and operating costs are low, purchasing a MALDI-TOF MS instrument requires a large initial investment.

IMMUNOLOGICAL METHODS

Serological determination of group-specific carbohydrate antigens of the cell wall has remained an important routine identification technique for beta-hemolytic streptococci since it was established by Rebecca Lancefield in the 1930s. Kits for grouping streptococci are based on latex agglutination or coagglutination testing and are readily available from different commercial suppliers. Streptococcal cells (after a rapid antigen extraction process) are mixed with latex particles or nonviable staphylococci coated with a group-specific hyperimmune antiserum. Positive reactions are visualized by clumping of the latex particles or staphylococcal cells. Because of their simplicity, rapidity, and specificity, agglutination tests are very practical methods and are widely used for routine diagnostic purposes. *S. pseudoporcinus* cross-reacts with GBS grouping antisera and may be misidentified as GBS (20, 21), although the incidence and importance of this recent finding are still under investigation.

DIRECT ANTIGEN DETECTION OF GBS

Several different antigen tests have been developed to detect GBS directly from vaginorectal samples. However, none of the currently available tests has an acceptable degree of sensitivity to detect GBS colonization in pregnant women, and the tests cannot be recommended for this purpose (4). In bacterial meningitis, antigen detection is not a substitute for a thorough bacterial examination of cerebrospinal fluid (CSF), because antigen testing is hampered by low sensitivity and specificity. Thus, a negative test result does not rule out infection caused by a specific meningeal pathogen. Although these tests can help in the diagnosis of neonatal GBS meningitis when no visible or viable microorganisms are present, the routine use of latex agglutination testing for the etiological diagnosis of bacterial meningitis has been questioned (1, 48).

MOLECULAR METHODS FOR DETECTION OF GBS

The development of molecular methods for specific diagnostic purposes has changed clinical microbiology considerably in recent years. In most cases, simple species diagnosis of GBS does not require NAATs. If atypical GBS isolates are encountered, however, a correct and reliable species designation can easily be achieved through sequencing of the 16S rRNA gene and determination of the *sodA* gene sequence. Much more potential for the application of NAATs in GBS diagnostics lies within GBS screening. A first PCR for GBS screening purposes was developed based on the CAMP factor gene (42). Several specific PCR assays for the detection of GBS colonization, employing different gene targets, have been developed and evaluated in recent years. Of those

tests, six have received U.S. Food and Drug Administration (FDA) clearance. Most of those NAATs are currently incorporated into automated molecular platforms such as the BD MAX system (Becton, Dickinson), the SmartCycler and Xpert technology (Cepheid), and the Illumigene system (Meridian Bioscience). Three of the tests are conducted directly with clinical swabs, while the other assays require a culture enrichment step prior to amplification. A current list of nucleic acid-based tests that have been approved by the FDA can be accessed at http://www.fda.gov/medicaldevices/productsandmedicalprocedures/invitrodiagnostics/ucm330711.htm. The main advantage of PCR is the short time frame necessary to provide results regarding GBS colonization status at the time of delivery. However, due to lower sensitivity, in comparison with cultural enrichment, the latest CDC guidelines for GBS prevention (4) recommend antenatal *S. agalactiae* screening by NAATs only after prior cultural enrichment in selective broth. Moreover, it must be noted that even a PCR method with a very short time frame to results may not provide results fast enough for sufficient administration of peripartum antibiotics, since many women deliver within a few hours after hospital admission. A further disadvantage is the lack of information on antibiotic susceptibility, which is required for all women reporting an allergy to penicillin. Therefore, the CDC currently does not recommend routine GBS screening by NAATs at delivery (4).

CONCLUSIONS

Despite the introduction of MALDI-TOF MS and rapid molecular methods, such as real-time PCR assays, that are now commercially available, the investment and expertise required limit their use in most laboratories. Therefore, many clinical microbiology laboratories rely on more traditional phenotypic methods for the identification of GBS.

The sensitivity of screening methods based on the culture identification of maternal carriage of GBS depends on the timing of specimen collection, the source of the specimen, and the culture technique used by the microbiology laboratory. The use of Granada-type and chromogenic media is a good alternative for the identification of GBS carrier status among near-term pregnant women. Moreover, culture methods for GBS detection remain a compulsory step for women reporting an allergy to penicillin.

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