Evaluation of a New Chromogenic Medium (StrepB Select) for Detection of Group B Streptococcus from Vaginal-Rectal Specimens

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We compared StrepB Select medium (Select) after enrichment with conventional culture for the detection of Group B Streptococcus (GBS). Postenrichment sensitivities of Select and conventional culture were 98.8% and 92.2%, respectively (P < 0.05). Select was superior for detection of GBS from vaginal-rectal specimens. Growth of non-GBS colonies required additional work to exclude the presence of GBS, especially after 48 h of incubation. Incubation of Select beyond 24 h did not significantly increase the yield of GBS.

As group B Streptococcus (GBS) remains a significant cause of neonatal morbidity and mortality, antenatal screening for GBS at 35 to 37 weeks of gestation is recommended to determine whether antimicrobial prophylaxis is warranted (4, 7). The use of newer, chromogenic media may improve the yield of GBS, while reducing labor and turnaround time (5). We evaluated a new chromogenic medium, StrepB Select (Select; Bio-Rad Laboratories, Marnes-la-Coquette, France), a selective medium for the detection and presumptive identification of GBS in vaginal and vaginal-rectal specimens (9). We compared the recovery of GBS from StrepB Select with that from conventional culture on colistin-nalidixic acid agar with 5% sheep blood (CNA; Oxoid, Nepean, Ontario, Canada) with and without broth enrichment using Streptococcus selective broth (SSB; Bio-Media Ltd., Woodbridge, Ontario, Canada).

From September to November 2008, a total of 1,025 specimens from 992 patients were submitted for GBS screening. These swabs were directly inoculated onto CNA plates and then placed into enrichment broth (SSB). After 24 h of incubation, the direct CNA plate was examined for the presence of colonies suggestive of GBS. If direct culture on CNA did not yield GBS, the SSB was subcultured onto CNA and incubated for 24 h. All broths were subcultured onto StrepB Select and incubated for up to 48 h at 37°C in ambient air, per the manufacturer’s recommendations. Colonies suggestive of GBS, with a turquoise blue color on Select and gray colonies with or without hemolysis on CNA, were worked up by separate, experienced technologists blinded to each other’s work. Identification of GBS was performed using conventional tests, including those with catalase, Gram stain, and Lancefield grouping antisera by using the PathoDx latex agglutination kit (Remel, Inc., Lenexa, KS). PCR testing for the cfb gene encoding the Christie-Atkins-Munch-Petersen (CAMP) factor (3), directly from the SSB enrichment broth, was used as the gold standard. A true positive was defined as growth of GBS on either medium.

Of the 1,025 specimens tested, a total of 243 (23.7%) yielded GBS, and the same 243 specimens were also positive by PCR for the cfb gene. Direct culture onto CNA yielded GBS from 201 samples (82.7%) (Table 1). SSB enrichment with CNA subculture at 24 h detected an additional 23 isolates (224/243 samples; 92.2%), while SSB enrichment with Select subculture detected GBS in 240 of 243 samples, for a sensitivity of 98.8% (P < 0.0001). There were no specimens that yielded GBS on direct culture that failed to grow in SSB. At 24 h postenrichment with Select, there were 50 specimens (6.4%) which grew turquoise blue colonies that were not GBS, the majority of which were Enterococcus spp. (n = 35) and Streptococcus bovis (n = 15). One specimen grew turquoise blue colonies on Select at 24 h postenrichment and tested positive for group B with PathoDx. Retrospective examination of the CNA plate for this specimen showed colonies with a large zone of beta-hemolysis, not characteristic of GBS. PCR for the cfb gene was negative, and 16S rRNA gene sequencing followed by BLAST analysis (National Center for Biotechnology Information [NCBI])

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<tr>
<th>Condition</th>
<th>No. (%) of samples positive for GBS</th>
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<tr>
<td>Preenrichment (CNA direct; 24 h)*</td>
<td>201 (82.7)</td>
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<tr>
<td>Postenrichment (SSB enrichment with CNA subculture, and SSB enrichment with StrepB Select subculture)</td>
<td>224 (92.2)</td>
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<tr>
<td>StrepB Select</td>
<td>24 h*</td>
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<tr>
<td>48 h*</td>
<td>241 (99.2)</td>
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| * | Samples inoculated directly onto colistin-nalidixic acid blood agar (CNA). |
| --- | "Postenrichment with Streptococcus selective broth followed by incubation onto colistin-nalidixic acid blood agar and incubated for 24 h. "Postenrichment with Streptococcus selective broth followed by incubation onto StrepB Select agar and incubated for 24 h. "Postenrichment StrepB Select agar incubated for 48 h. The total number of GBS-positive specimens was 243. |
identified the isolate as *Streptococcus pseudoporcinus* (100% identity) (2). Incubation of *Select* plates for an additional 24 h yielded only one other specimen with GBS, for an overall sensitivity of 99.2%. There were 294 specimens (37.5%) which had blue colonies that were not confirmed as GBS (Fig. 1) at 48 h. *Select* recovered 17 more isolates following SSB enrichment than CNA. *Select* failed to detect GBS from two specimens that grew on CNA, as the colonies on *Select* agar were white in color. PCR performed from these white colonies confirmed the isolates as GBS.

Current guidelines for prenatal GBS screening recommend obtaining a vaginal-rectal swab and the use of selective broth enrichment, to maximize sensitivity (1, 7). Few studies have examined the use of chromogenic media for the detection of GBS from screening specimens (6, 8, 9). In this evaluation, although StrepB *Select* medium demonstrated excellent sensitivity for the detection of GBS, there are a few limitations that need to be mentioned. Because of the chromogenic substrates present, storage and incubation of the media must be in the dark, and minimal exposure to light is necessary for optimal performance. A number of non-GBS organisms, such as *Enterococcus* spp., group A *Streptococcus*, *Streptococcus bovis*, and *Streptococcus pseudoporcinus*, may grow as turquoise blue colonies. There are also rare strains of GBS that may produce no activity for the detection of GBS, the identification of suspicious colonies must be confirmed by conventional testing. A minimally increased yield of GBS was realized by prolonging the incubation of *Select* plates beyond 24 h following broth enrichment, but there was a significant increase in the work required to confirm the identity of non-GBS isolates that had a blue color. These were all *Enterococcus* spp. (bile esculin positive) that were purple at 24 h but turned blue at 48 h (Fig. 1c). One limitation of this study was that no comparison of postenrichment CNA subculture after 48 h incubation was made. However, in a study by Smith et al., no significant difference in sensitivity was noted between the 24 and 48 h results on CNA or sheep blood agar after enrichment (8).

We thank Bio-Rad Canada for providing the StrepB *Select* medium for this evaluation.

**REFERENCES**