Abstract

Group B Streptococcus (GBS) remains one of the leading causes of sepsis and meningitis in newborns despite recent advances in the prevention of neonatal group B streptococcal (GBS) disease. Although the current gold standard method is the LIM broth method as recommended by the Centers for Disease Control and Prevention (CDC), several alternative methods such as StrepB Carrot Broth™ and Granada™ broth have been developed with comparable specificity, sensitivity, and reduced turnaround time. The goal of this study was to evaluate and compare the newly commercialized method - StrepB Carrot Broth™ against the LIM broth protocol.

Results

Introduction

GBS and GAS reference strains and Eighteen clinical isolates were tested with the two methods and three dilutions (1.5 x 10^4 CFU/mL, 1.5 x 10^3 CFU/mL, 1.5 x 10^2 CFU/mL) for evaluating the detection of limit. 100 clinical specimens were collected and tested in parallel by both StrepB Carrot Broth™ and traditional LIM broth protocols. Both the methods presented 100% sensitivities in dilutions of 1.5 x 10^4 CFU/mL, 1.5 x 10^3 CFU/mL, and 1.5 x 10^2 CFU/mL. At a concentration of 1.5 x 10^2 CFU/mL, StrepB Carrot Broth™ was able to recover GBS in at least 19 samples, while LIM Broth was able to cover GBS in 15 of 19 samples. 20 (20.0%) clinical specimens were detected as positive for GBS by StrepB Carrot Broth™ and 19 (19.0%) by LIM Broth enrichment protocols while direct plating only detected 13.

Based on the evaluation, the StrepB Carrot Broth™ 24 hour broth method was shown to be more sensitive comparing to LIM broth and has equivalent clinical performance against LIM broth protocol. Besides, the application of the StrepB Carrot Broth™ can shorten the time to identify beta haemolytic GBS from at least 48 hours to 24 hours without further identification testing such as streptococcal grouping latex agglutination test, presumptive identification testing by CAMP test, and etc. Strep B Carrot Broth™ can be employed as reliable method for detection of beta-haemolytic Group B Streptococci.

Materials and Methods

Sample Preparation and Inoculation

Stage I

1. A Group B streptococci isolates and reference strains of Group B Streptococcus and Group A Streptococcus were analyzed in this evaluation.
2. A suspension equivalent to 0.5 McFarland was prepared for each isolate. Serial dilutions of 1.5 x 10^4 CFU/mL, 1.5 x 10^3 CFU/mL, and 1.5 x 10^2 CFU/mL, was prepared.
3. An amount of 100 µL of each dilution was inoculated to StrepB Carrot Broth™ and LIM Broth.
4. StrepB Carrot Broth™ were inoculated, incubated, and interpreted according to manufacturer’s recommendations.

Stage II

1. 100 clinical GBS screening specimens were collected since June 1, 2011.
2. Specimens were collected using swabs, inoculated on blood agar plate, and vortexed for 30 seconds. The suspension was evenly spotted into StrepB Carrot Broth™ and LIM Broth.
3. StrepB Carrot Broth™ and LIM Broth were then subcultured on to blood agar plate after overnight incubation.

Interpretation

StrepB Carrot Broth™

1. All tubes that turned orange to red were considered to be positive.
2. All tubes that did not turn orange to red color were considered as negative.

LIM Broth

1. Samples that yielded GBS after subculture to blood agar plate and identified by latex assay for serotyping were considered as positives.
2. Samples that did not yield GBS after subculture to blood agar plate were considered as negatives.

Discussion

1. Based on the Stage I findings, StrepB Carrot Broth™ are very reliable in GBS detection considering the fact that most clinical specimens, when positive for GBS, will most likely have a GBS load above 10^3 CFU/mL.
2. In simulations of low GBS count, StrepB Carrot Broth™ was more sensitive than LIM Broth in this evaluation.

GBS Positive

<table>
<thead>
<tr>
<th>CFU/mL</th>
<th>Group B Streptococci Recovery Rate</th>
<th>LIM Broth</th>
</tr>
</thead>
<tbody>
<tr>
<td>10^1</td>
<td>100% (19/19)</td>
<td>100% (19/19)</td>
</tr>
<tr>
<td>10^2</td>
<td>100% (19/19)</td>
<td>100% (19/19)</td>
</tr>
<tr>
<td>10^3</td>
<td>84% (16/19)</td>
<td>79% (15/19)</td>
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GBS Negative

<table>
<thead>
<tr>
<th>CFU/mL</th>
<th>Group B Streptococci Recovery Rate</th>
<th>LIM Broth</th>
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