

Evaluation of StrepB Carrot Broth™ and LIM Broth for Long Term Storage/Recovery of Group B Streptococcus

J.R. DiPersio, J. Beach and L. Define
Summa Health System, Akron, OH

Abstract

Background: 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 disease. The current CDC recommended OBGYN screening method involves use of a selective broth such as LIM or Trans-Vag, but several alternative methods such as StrepB Carrot Broth™ have been developed with comparable sensitivity, specificity, and reduced turnaround time.

Purpose: This study evaluated the ability of StrepB Carrot Broth™ and standard LIM Broth to maintain the viability of GBS over time following specimen inoculation. Use of these broths as a storage medium would facilitate antimicrobial susceptibility testing at a later date if penicillin-allergic patients are not identified at the time of culture.

Methods: One hundred vaginal/rectal patient specimens were cultured in parallel using both broth methods. Each broth system detected the same 21 GBS positive patients (using manufacturers' recommended procedures). Positive broths were held at room temperature and further evaluated for long-term GBS recovery by subculturing onto SBA for semi-quantitation of growth.

Results: GBS were recovered from either broth system for a minimum of 13 days and usually much longer. GBS survival in StrepB Carrot Broth™ averaged 51 days (range 15 to 113 days) and GBS survival in LIM Broth averaged 32 days (range 13 to 90 days). GBS survival in StrepB Carrot Broth™ was longer than in LIM broth for 19 of 21 positive specimens. In 2 of 21 specimens, GBS survived longer in LIM Broth.

Conclusion: Both broth systems maintained the viability of GBS at room temperature for at least 13 days and usually much longer, but StrepB Carrot Broth™ maintained viability longer than LIM broth for most GBS isolates. Use of these broths as a storage medium should allow adequate time for follow-up testing when needed.

Introduction

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 disease. Approximately 10-30% of pregnant women are asymptomatic carriers of GBS, which represents a risk to the newborn. The CDC currently recommends OBGYN screening for intrapartum GBS colonization at 35–37 weeks gestation. Culture based methods involve the inoculation of vaginal/rectal specimens into a selective broth such as LIM or Trans-Vag, followed by subculture onto blood agar. Several alternative methods such as StrepB Carrot Broth™ have been developed with comparable sensitivity, specificity, and reduced turnaround time. Newer molecular methods of detection are also now in use for this purpose. Identification of women colonized with GBS around the time of delivery allows physicians to better assess the need for prophylactic antimicrobial treatment.

Penicillin is the preferred agent for use against GBS and confirmed resistance has not been observed to date. Anaphylaxis associated with GBS prophylaxis is rare but alternative treatment with a non-beta lactam antibiotic may be warranted in penicillin-allergic women at high risk. Macrolide and lincosamide class drugs may be used as alternatives to Penicillin but in-vitro antimicrobial susceptibility testing (AST) of GBS is recommended due to rising rates of resistance. Some laboratories perform routine AST on all GBS while others perform testing only when Penicillin allergy is noted at the time of specimen processing. If AST is not routinely performed on all GBS isolates, the laboratory should hold isolates for a reasonable amount of time for possible additional testing requests even after culture results are finalized.

The purpose of this study was to evaluate the ability of StrepB Carrot Broth™ and standard LIM Broth to maintain the viability of GBS over time following specimen inoculation. Use of these broths as a storage/holding medium would facilitate antimicrobial susceptibility testing at a later date if penicillin-allergic patients are not identified at the time of culture.

Methods

Vaginal/rectal swabs were collected using the BBL CultureSwab™ (dual swab) Collection & Transport System (Becton Dickinson). One swab was used to inoculate a tube of LIM broth (BD) and the other to inoculate a tube of StrepB Carrot Broth™ (Hardy Diagnostics) following manufactures directions. One hundred vaginal/rectal patient specimens were cultured in parallel using both methods.

All LIM and StrepB Carrot Broth™ tubes were sub-cultured onto blood agar following incubation for up to 24 hrs at 35°C. Blood agar plates were streaked like a urine culture using a 0.001 ml loop; first down the center of the plate, followed by cross streaking. Plates were incubated at 35°C for 24-48 hrs.

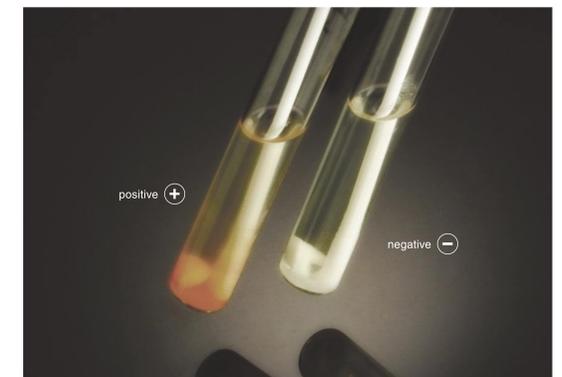
LIM and/or StrepB Carrot Broth™ tubes, initially positive for GBS were held at room temperature and sub-cultured onto blood agar at periodic intervals (usually 3x weekly) until two successive subcultures failed to yield identifiable GBS colonies. Suspected GBS colonies as well as normal flora colonies were semi-quantitated (no growth to 4+ growth).

Results

Each broth system detected the same 21 GBS positive patients out of the 101 patient specimens tested. Results of sequential subcultures are shown in table 1. Survival in LIM Broth ranged from 13 to 90 days (avg = 32 days) and survival in StrepB Carrot Broth™ ranged from 15 to 113 days (avg = 51 days). GBS survival in StrepB Carrot Broth™ was longer than in LIM broth for 19 of 21 positive patients.

Table 1. Recovery Rates of GBS from StrepB Carrot Broth™ and LIM Broth Over Time

Specimen Number	StrepB Carrot Broth™ Color Reaction	StrepB Carrot Broth™ Number of Days positive	Lim Broth Number of Days positive
1	1+	15	ND
2	2+	18	ND
3	2+	47	20
4	4+	32	24
5	1+	84	19
6	4+	42	37
7	4+	20	13
8	4+	41	13
9	3+	40	13
10	4+	54	22
11	4+	46	26
12	2+	53	90
13	2+	38	34
14	1+	77	26
15	2+	34	27
16	4+	113	27
17	3+	83	27
18	1+	39	19
19	4+	56	32
20	4+	109	49
21	3+	26	84
	Mean	51	32
	Range	15-113	13-90



Conclusions

- Both broth systems maintained the viability of GBS for at least 13 days at room temperature and usually much longer.
- StrepB Carrot Broth™ maintained viability much longer than LIM broth (average of 51 days vs. 32 days respectively).
- Use of either broth as a storage medium should allow adequate time for follow-up AST by the laboratory if needed.

References

- Schrag et al. (2002) Prevention of perinatal group B streptococcal disease. Revised guidelines from CDC. MMWR Recomm. Rep. 51:1-23.
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- DiPersio L, and J. DiPersio. 2006. High rates of erythromycin and clindamycin resistance among OBGYN isolates of group B *Streptococcus*. Diag. Microbiol. Infect.t Dis. 54:79-82.