University of Michigan Health System

Molecular vs. Culture: Detection of Group B Streptococcus (GBS) Colonization in Pregnancy

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Abstract

Background: In 2008, our laboratory implemented molecular testing (BD GeneOhmStrepB) (PCR) as an adjunct to LIM enriched culture for perinatal GBS screening. In 2010 the CDC screening guidelines endorsed use of PCR, following enrichment. We experienced periodic problems with inhibition of our PCR test which necessitated repeat testing and decided to evaluate other culture-based methods: Strep B Carrot Broth™ (Hardy Diagnostics, Santa Monica, CA) and GBS Detect™ (Detect) (Hardy).

Methods: Currently, vaginal/rectal swabs submitted for GBS screening are inoculated directly onto NEL-GBS (NEL, Northeast Labs, Winslow, ME) which is incubated anaerobically, then added to LIM broth (Remel, Lenexa, KS). If no orange colonies are detected on NEL at 24 hours, PCR is performed on LIM using BD GeneOhm StrepB (Franklin Lakes, NJ). If the PCR is positive, the LIM is subcultured to blood agar for susceptibility testing of the isolate. We compared 3 approaches for GBS detection: chromogenic detection of GBS (NEL and Carrot); PCR from LIM and Carrot; and subculture of LIM and Carrot to GBS Detect™ .

Results: One hundred and four vaginal/rectal swabs were tested, 24 were positive for GBS: NEL direct plate 17/24 (71%), Carrot broth 19/24 (79%); BD PCR 23/24 (96%), Detect 24/24 (100%).

Method		Number of positive	Sensitivity/ Specificity	Cost	Time/test (min)
Chromogenic	GBS, direct	17	71/100	X	0.1
	Carrot	19	79/100	X	0.1
PCR	LIM	23	96/100	13X	3.9
	Carrot*	24	100/96	13X	3.9
Enriched subculture	LIM/Detect	23	96/100	X	0.6
	Carrot/Detect	24	100/100	X	0.6

X = approximately $:*not FDA approved

Conclusions: PCR is more rapid than culture but 13 times more expensive and does not improve detection of GBS. Carrot broth detected more GBS than NEL and does not require anaerobic incubation. Subculture of all Carrot to Detect easily isolated GBS, including nonhemolytic GBS, for susceptibility testing which our clinicians require. We changed to Carrot broth followed by subculture to Detect which is more cost and time efficient in our academic health center and provides optimal reporting to our clinicians.

Introduction

The purpose of this study was to re-evaluate our current method for GBS screening cultures, direct plating to NEL-GBS and LIM, followed by real-time reverse transcriptase PCR (BD GeneOhm StrepB). We have periodic problems with inhibition in our PCR assay which requires repeat testing, adding time and cost to our assay. We decided to evaluate Carrot broth as a substitute for NEL-GBS and LIM and determine the performance characteristics of PCR from Carrot. This study sought to determine whether subculture of enrichment broth to GBS Detect, a newer media for the isolation of hemolytic and nonhemolytic GBS would be of value in our testing algorithm.

Method

Specimen: Vaginal/rectal swabs (n=104)	Plate Media	Broth Media
Primary	NEL-GBS (Northeast Labs)	LIM (Remel) Carrot (Hardy)
Incubation	18-24 hours, anaerobic	18-24 hours, CO_2
Read	Orange colonies → +	Carrot orange → +
PCR		GeneOhm StrepB (Becton Dickinson)
Subculture	GBS Detect	GBS Detect (Hardy)
Identification	CAMP test	CAMP test

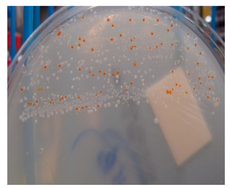
Results

Detection of GBS

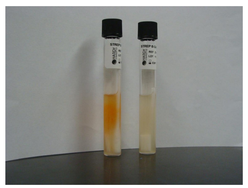
Detection	Media	Positive	Sensitivity/ Specificity	Unresolved
Chromogenic	NEL-GBS	17	71/100	
	Carrot	19	79/100	
Molecular (PCR)	LIM	23	96/100	2*
	Carrot	24	100/96	6*
Broth → GBS Detect	LIM	23	96/100	
	Carrot	24	100/100	

* Unresolved = PCR was inhibited

NEL-GBS



Carrot



GBS Detect

Cost and Time

Detection	Media	Cost/Test	Time/Test (minutes)
Chromogenic	NEL-GBS	X	0.1
	Carrot	X	0.1
Molecular (PCR)	LIM	13X	3.9
	Carrot	13X	3.9
Broth → GBS Detect	LIM	X	0.6
	Carrot	X	0.6

Culture (Carrot → GBS Detect) 0.7 minutes
PCR (LIM or Carrot) 3.9 minutes

Discussion

Twenty-four positive GBS results were detected by PCR and GBS Detect. One false positive PCR from Carrot was suspected to be caused by a reaction with *Streptococcus porcinus*, which was isolated from the culture. A false negative PCR and culture result from LIM was attributed to sampling error. Unresolved PCR results may be caused by inhibitors in the sample, method of broth inoculation, and inconsistencies in pipetting technique. GBS Detect agar has supplements that cause gamma and weakly hemolytic Strep B colonies to present large zones of hemolysis. Other organisms will cause this reaction to occur, but are CAMP negative. Our laboratory confirms all Carrot Broth negative, GBS Detect positive cultures with serologic typing or CAMP test.

Conclusions

- **Carrot broth reliably detects beta hemolytic GBS in 16-24 hours**
- **Subculture of Carrot broth to GBS Detect easily isolates GBS (hemolyic and nonhemolytic) for susceptibility testing**
- **Culture is 13 times less expensive than PCR**
- **Culture is takes less personnel time than PCR**