

Comparison of Culture Methods for the Detection of Group B Streptococcus from Vaginorectal Specimens

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Revised Abstract:

Group B Streptococcus (GBS) is an important pathogen in neonates. The Centre for Disease Control (CDC) revised guidelines in 2002 recommend screening women for GBS carriage at 35 to 37 weeks gestation by culture of a vaginorectal swab using a selective broth with subculture onto a plate. The objective of our study was to compare our current method (selective broth with subculture) with others available so as to improve the sensitivity of detection of GBS in our laboratory.

Method

Vaginorectal swabs (200) were screened for GBS using direct inoculation to 5% sheep blood and SXT (BA + SXT) (PML); swab was vortexed in 2mL of Tryptic soy broth and inoculated into Group B Selective Broth and StrepB Carrot Broth (Hardy Diagnostics). Following incubation the broths were tested with Prolex Streptococcal latex agglutination (LA) (Prolab), subcultured onto Tryptic soy agar with 5% sheep blood (BA) (PML) and onto Granada Medium (Hardy Diagnostics). Plates were examined at 24 and 48 hrs.

	Direct Plate	BA Subculture	Granada Medium Subculture	GBS Selective Broth LA	StrepB Carrot Broth LA
			Subculture	DIOIII LA	DIOIII LA
Total number tested	200	200	200	63	63
Sensitivity (%)	71	86	89	77	100
Specificity (%)	99	83	96	98	98

Conclusio

The sensitivity of direct plate was low. The agglutination tests direct from the Group B Strep broth improved sensitivity, but the specificity was low. Granada medium showed improved specificity compared to BA plates. Preliminary data using StrepB Carrot broth indicated both improved sensitivity and specificity. Continued testing with this media is on-going and if preliminary findings are confirmed, this media will be incorporated into our routine for GBS screening.

Background

Group B Streptococcus (GBS) is one of the leading causes of infectious morbidity and mortality in neonates in North America. The colonization rate with GBS in pregnant women is between 15 – 30%. The Centre for Disease Control (CDC) guidelines published in 2002 recommend screening women for GBS carriage at 35 to 37 weeks gestation, and use intrapartum antibiotics in positive mothers as the first line approach in the prevention of neonatal GBS sepsis. The guidelines recommend the use of a vaginorectal swab to be placed into a selective broth with subculture onto a Blood Agar plate or other appropriate media. Several alternative methods have been developed which may increase the sensitivity of the detection of GBS.

Objective

The objective of our study was to compare our current method (selective broth with subculture) with the addition of Carrot Broth, Granada agar, and Latex Agglutination to determine if we could improve the sensitivity of detection of GBS in our laboratory.

Method:

Vaginorectal swabs submitted for GBS screening were processed as follows:

	Media	Culture Method	Incubation / Reading
1)	Tryptic soy agar plate with 5% sheep blood and SXT (BA + SXT) (PML)	Swab inoculated directly onto plate	Plate incubated at 35°C CO ₂ , then examined for GBS at 24 and 48 hours incubation
2)	Group B Streptococcus Selective Broth (PML)	Swab vortexed in 2mL of TSB. 0.5mL inoculated into Group B Selective Broth	Broth incubated at 35°C CO ₂ for 18 to 24 hours incubation a) then tested with the Prolex Streptococcal agglutination (Prolab), and b) subcultured onto Tryptic soy agar with 5% sheep blood (BA) (PML) and c) Granada Medium (Hardy Diagnostics). Plates examined for GBS at 24 and 48 hours incubation.
3)	StrepB Carrot Broth Kit (Hardy Diagnostics)	Swab vortexed in 2mL of TSB. 0.5mL inoculated into into StrepB Carrot Broth	Broth incubated at 35°C O ₂ for 18 to 24 hours, and a) examined for the production of an orange color, b) then tested with the Prolex Streptococcal agglutination (Prolab), and c) subcultured onto BA, and d) Granada Medium. Plates examined for GBS at 24 and 48 hours incubation.

Methods 1, 2, and 2a comprise our current method.

Results:

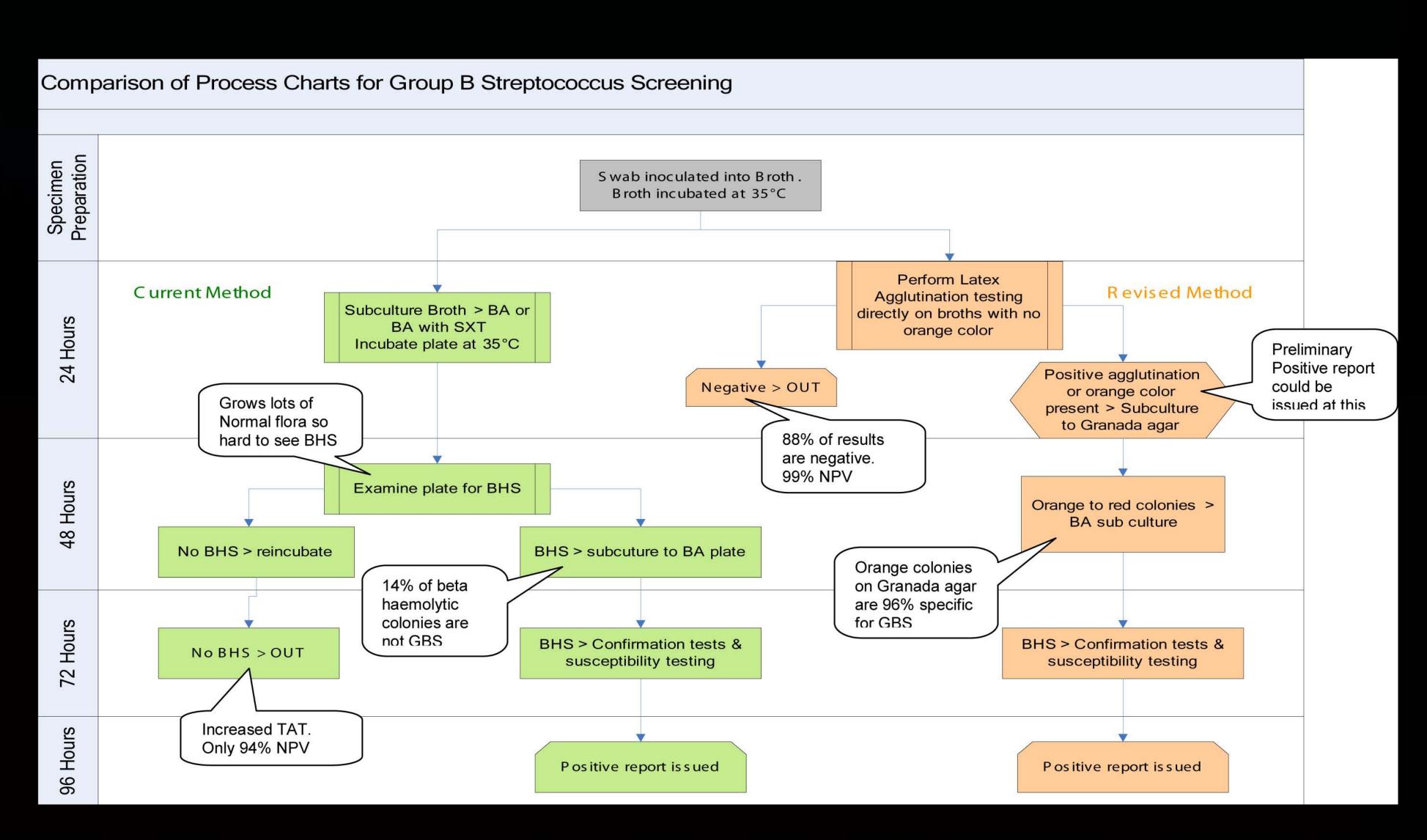
	BA + SXT Direct Plate	BA Sub	Granada Medium Sub	Group B Strep Broth tested by latex agglutination	StrepB Carrot Broth tested by latex agglutination	Strep B Carrot Broth orange color
Total number tested	200	200	200	63	63	63
Sensitivity (%)	71	86	89	77	100	92
Specificity (%)	99	83	96	98	98	100
PPV (%)	94	56	87	91	92	100
NPV (%)	92	96	97	94	100	98

Results (continued):

- 1. The use of the direct plate improved the TAT, but the method has the lowest sensitivity.
- 2. Subculture onto a selective agar or BA had the same sensitivity, however, the specificity was higher with the use of the selective agar (96%)
- 3. The sensitivity of latex agglutination performed directly from the selective broth increased from 77% to 100% when done from the STREPB Carrot broth.
- 4. Another advantage of the STREPB Carrot broth was the colour change (92% sensitive).

Conclusion:

Optimal results in this comparison study were found with the use of selective STREPB Carrot Broth and Granada agar. Adopting this change would improve the sensitivity, specificity, NPV and TAT compared to our current method.



References

- 1. Schrag S, Zywicki S, Farley M, et al. Group B streptococcal disease in the era of intrapartum antibiotic prophylaxis. N Engl J Med 2000; 342: 15-20.
- 2. Regan JA, Klebanoff MA, Nugent RP, Vaginal Infections and Prematurity Study Group. The epidemiology of group B streptococcal colonization in pregnancy. Obstet Gynecol 1991; 77: 604-10.
- Centers for Disease Control and Prevention, 2002, Prevention of perinatal group B streptococcal disease: revised guidelines from CDC. Recommendation and Reports.
 H. Isenberg, Clinical Microbiology
- H. Isenberg, Clinical Microbiology
 Procedures Handbook, Group B
 Streptococcus Cultures, Vol. A, Section 3,
 2004, ASM Press, Washington, DC.

 F. Baron, Laboratory Support for Prevention
- 5. E. Baron, Laboratory Support for Prevention of Perinatal Group B Streptococcal Disease: Commentary on the New Guidelines on Screening for Group B Streptococci During Pregnancy, Clinical Microbiology Newsletter, Vol. 25, No. 9, May 1, 2003.
- 6. Park, C., et al, 2001. Detection of Group B Streptococcal Colonization in Pregnant Women Using Direct Latex Agglutination Testing of Selective Broth
- 7. StrepB Carrot Broth Kit, Product Literature, Hardy Diagnostics.
- 8. Granada Medium, Product Literature, Hardy Diagnostics.

