Evaluation of A New Chromogenic Group B *Streptococcus* Agar and Use of Digital Analysis for Detection of Group B *Streptococcus* in Vaginal/Rectal Swabs

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ABSTRACT

Detection of Group *B Streptococcus* (GBS) colonization during pregnancy aids in the prevention of early onset GBS disease in newborns. A new Group B chromogenic culture medium combined with segregation software (SSW) developed for the WASPLab™ system (Copan Diagnostics) may aid in the accurate detection of GBS colonization as well as enhanced laboratory workflow management. We evaluated the LIM broth/ChromID® Strepto B agar (STRB, bioMerieux) for detection of GBS along with the WASPLab SSW for automated digital analysis for separation of negative and positive culture results. The comparative method was Carrot broth/GBS Detect medium (Hardy Diagnostics). Vaginal/rectal swabs were processed by the WASPLab system using both methods with initial inoculation into the enhancement broths which were incubated off-line at 35-37°C overnight, followed by WASP inoculation of the plates and incubation at 35-37°C for 20 h (Detect) and 24 h (STRB). WASPLab digital images of GBS Detect (beta hemolysis) and STRB (pink to red colonies considered positive) were visually examined. STRB also had digital images analyzed by the SSW to automatically segregate negatives from positives. All positives were confirmed as GBS or not GBS by phenotypic methods. There were 245 samples acceptable for comparison with 152 negative by Detect and STRB and 87 positive by both methods. There were 4 cultures positive by Detect that were negative by STRB and 2 cultures that were negative by Detect and positive by STRB. Sensitivity was 96% and specificity 99%, however the adjusted specificity was 100% with the 2 STRB positive/ Detect negative cultures considered to be true positives. The 245 STRB plate images were analyzed by SSW after initial visual examination. There were 89 STRB visually examined and determined to be positive with 100% detected as positive by SSW with no false negatives. Of the remaining 156 STRB negative plates, 124 (79%) were negative by SSW. There were 32 positives indicated by SSW that were determined to be negative for GBS by additional phenotypic testing. These were determined to be SSW false positive most likely due to blue/purple colonies with a pink hue in the medium. The STRB is equivalent to the Detect for detection of GBS. The SSW is a powerful tool for automatic separation of positive and negative cultures to help laboratory workflow.

INTRODUCTION

Streptococcus agalactiae is a colonizing microorganism in pregnant women. Genitourinary tract vaginal colonization usually occurs in late adolescence. Women of child-bearing age carry GBS at variable frequencies once colonized. The rate of GBS colonization among pregnant women usually remains stable over time. Additionally, colonization toward the end of pregnancy is a risk factor for potentially severe newborn diseases, including neonatal sepsis.

GBS infection in newborns arises by the aspiration of infected amniotic fluid or vertical transmission during delivery through the birth canal. Due to the high neonatal mortality rate due to maternal GBS colonization, the Centers for Disease Control and Prevention recommended universal *intrapartum* antimicrobial prophylaxis in women of high risk at 35-37 weeks of gestation and screening of anorectal and vaginal specimens at 35-37 weeks of pregnancy is recommended. In this study, we evaluated STRB for the qualitative detection of GBS in pregnant women and utilized the Copan WASPLAB digital analysis software for detection of GBS and segregation of positive from negative cultures.

METHODS

A total of 245 Vaginal/Rectal swabs were processed on one WASP line with incubation at 35-37^{0}C, non-CO_2 in a WASPLAB incubator.

1. WASP instrument was used to inoculate 30µL of Eswab specimen to a LIM broth. The inoculated LIM broth was removed and incubated off-line at 35-37^{0}C, in non-CO_2.
2. After 18-24 hours incubation, 30µL of incubated LIM broth was plated by the WASP to a STRB chromogenic agar plate.
3. Plates were placed in a WASPLab incubator in non-CO_2 at 35-37°C.
4. At 0 hours and after 24 hours incubation, digital imaging of all plates was performed within the WASPLab.
   a. All images were viewed by a CLS with positive plates removed for organism identification confirmation according to manufacturer's protocols and compared to laboratory results with Carrot Broth/Group B Detect.
   b. All plate images of STRB were also evaluated by Copan segregation software (SSW) which may be used as a preliminary screen to segregate negative from positive plates. Image analysis results were compared to manual CLS read results.
      a. Images of all plates were compared to results provided by the CLS read manual method.
      b. Discrepant results were subject to image review of discrepancies by a manager/director.
      c. Discrepancies of SSW positive, CLS negative were not considered significant. SSW would direct CLS to manually examine plate for further workup.

RESULTS

1. 245 total GBS cultures were evaluated by a CLS and by the SSW. See Tables 1 and 2.
2. There were 152 GBS cultures considered Negative and 87 Positive by both Detect and STRB .
   a. 4 cultures positive by Detect and negative by STRB
   b. 2 cultures negative by Detect and positive by STRB
   See Table 1
3. There were 245 STRB plate images analyzed by SSW after initial visual examination.
   a. 89 STRB positives by both visual examination and SSW.
      i. Zero were determined negative by SSW that grew GBS in culture.
   b. Among 156 negative STRB by visual examination,
      i. 124 (79%) were negative by SSW.
      ii. 32 were determined positive by SSW.

TABLE 1. Correlation between Detect and STRB

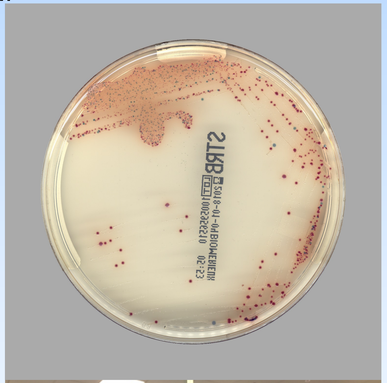
		STRB	
		Negative	Positive
GBS Detect Medium	Negative	152	2
	Positive	4	87

TABLE 2. Correlation between Visual examination and SSW

		SSW	
		Negative	Positive
Visual Examination	Negative	124	32
	Positive	0	89

CONCLUSIONS

1. The STRB is equivalent to the Detect for recovery and isolation of GBS.
2. The SSW is a powerful tool for automatic separation of positive and negative cultures to help laboratory workflow.



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