18th ECCMID Barcelona 19–22 April 2008
ESCMID EUROPEAN SOCIETY OF CLINICAL MICROBIOLOGY AND INFECTIOUS DISEASES

P 1388

EVALUATION OF THE STREPB SELECT AGAR FOR THE DETECTION OF GROUP B STREPTOCOCCI FROM VAGINAL AND RECTO-VAGINAL SPECIMENS

P. Melin, P. Huynen, C. Meex, P. De Mol

Natl. Reference Lab. for GBS, Univ. Hosp. of Liège, Liège, Belgium

Medical Microbiology, CHU, B-23
Sart Tilman - B 4000 Liège, BELGIUM
Phone: +32-4 366 24 38
Fax: +32-4 366 24 40
Email: Pierrette.Melin@chu.ulg.ac.be

UNIVERSITÉ de Liège

ABSTRACT

Background: Current guidelines for the prevention of group B streptococcal (GBS) disease are based on prenatal screening culture for recto-vaginal GBS colonization. Use of selective and differential media as Granada type agar (GRA) or ChromID StreptoB agar (SBID) improves sensitivity and workload of these cultures. This study was conducted to evaluate performances of the new StrepBSelect Agar (SBS) Bio-Rad, for the selective growth of blue-turquoise colonies of GBS.

Methods: 500 genital swabs collected from pregnant and non-pregnant women. Each swab was homogenized in 2 ml of sterile saline and 0.05 ml aliquots were inoculated onto SBS, modified GRA (Becton Dickinson), SBID (bioMerieux) and blood agar with colistin-nalidixic acid (CNA), primary cultures. The remaining suspension was added to a selective enrichment Lim broth. After overnight incubation, aliquots of Lim broth were inoculated onto SBS, GRA, SBID and CNA. SBS and SBID were incubated in air, GRA anaerobically and CNA in air + 7% CO2, at 35°C, 24 - 48 h. Positive and negative control strains (GBS; *E.faecalis*) were cultured with each run. Specific identification of colonies suggestive of GBS (light blue to dark blue-turquoise on SBS, light pink to red on SBID, beta-hemolytic on CNA) was performed; orange colonies on GRA were identified as GBS.

Results: GBS were recovered from 147 swabs (29.4 %): 111 from primary cultures and 139 after Lim enrichment, respectively from 103 and 134 on SBS, 90 and 123 on GRA, 93 and 124 on SBID, 76 and 113 on CNA. Overall sensitivities were 94.6% on SBS, 84.4% on GRA, 87.1% on SBID and 81.6% on CNA. Characteristic colonies of presumptive GBS were not always confirmed as GBS: 41 from primary cultures and 38 after Lim enrichment on SBS, 22 and 17 on SBID and 45 and 59 on CNA. Respectively the positive predictive values of presumptive GBS colonies were 71.5-77.9% (SBS), 80.9-87.9% (SBID) and 62.8-65.7% (CNA). At 48h incubation, presumptive GBS were easily observed on SBS, GRA and SBID even in low numbers.

Conclusions: 1) The highest sensitivity was observed for SBS, followed by these on SBID and GRA. 2) Due to lack of specificity, characteristic colonies on SBS as on SBID or CNA must be isolated to confirm their identification 3) Presumptive GBS were easily observed on SBS, GRA and SBID. 4) SBS as SBID are incubated in air and do not require CO_2 or anaerobic conditions. 5) SBS, a new useful agar to recommend for GBS prenatal screening culture.

BACKGROUND

To prevent GBS perinatal diseases, current guidelines recommend intrapartum antibioprophylaxis for women "at risk"; they are based on prenatal screening culture of all pregnant women at 35-37 weeks of gestation for rectal and vaginal GBS colonization. To provide the highest sensitivity, culture methods must include an enrichment in selective broth like Lim broth, further sub-cultured on a blood agar plate. However, this enrichment broth is not totally selective for GBS and other Gram positive cocci may as well be enriched by this method, possibly hiding GBS.

Use of selective and differential media as Granada type agar or ChromID StreptoB agar improves sensitivity and workload of these cultures.

OBJECTIVE

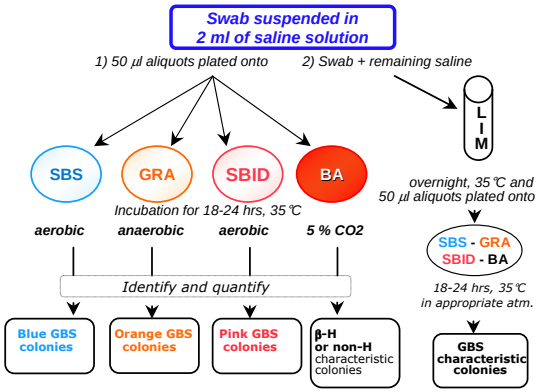
◆**To evaluate the performance of the StreptoB Select Agar (SBS), a new chromogenic medium, Biorad France, for the selective growth and identification of β-hemolytic (β-H) and non-hemolytic (H) GBS as blue-turquoise colonies.**

◆**By comparison with culture on Granada (GRA), StrepB ID (SBID) and Columbia sheep blood agar with colistin/ nalidixic acid (BA).**

METHODS

Clinical specimens and culture procedure

From August to November 2007, a total of 500 vaginal or vagino-rectal swabs were collected from pregnant or non-pregnant women at the university hospital of Liege.



If negative, plates were re-incubated overnight in appropriate atmosphere and inspected again at 48 hrs.
All plates were read independently from each other.

***Presumptive GBS colonies confirmed** with latex agglutination test*

A positive, GBS, and negative, *Enterococcus faecalis*, control strains were cultured with each run.

Culture Media

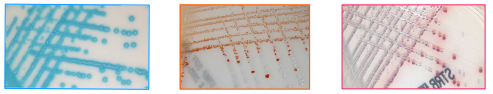
SBS, Strep B Select, Biorad, pilot batch
GRA, Granada - Strep B Differential Agar, Becton Dickinson
SBID, ChromID Strepto B Agar, bioMerieux
BA, Columbia Sheep Blood agar with Colistin/nalidixic acid, Biorad
LIM, Lim Broth (Todd Hewitt broth with colistin/nalidixic acid), Becton Dickinson

RESULTS

GBS Cultures (48 h)

On these 3 selective differential media, colonies were usually tiny after overnight incubation. The easiest to read after overnight incubation is the Granada agar, but a prolonged incubation is needed for all 3 agars.

GBS on StrepB Select *GBS on Granada* *GBS on Strep B ID*



GBS colonization rate

Overall, the colonization rate was **29.4%:** GBS were isolated from 147/500 swabs. Two isolates were non-hemolytic strains.

Sensitivity to detect GBS

Detection of GBS from vaginal/vagino-rectal swabs

	Positive cultures: number (% of positive swabs)				
Cultures	**SBS**	**GRA**	**SBID**	**BA**	**Overall**
Direct	103 (**70.1**)*	90 (**61.2**)	93 (**63.2**)	76 (**51.7**)	111 (**75.6**)
After Lim	134 (**91.1**)	123 (**83.7**)	124 (**84.3**)	113 (**76.9**)	139 (**94.6**)
Overall	139 (**94.6**)	124 (**84.4**)	128 (**87.1**)	120 (**81.6**)	**147**

*: SBS is significantly more sensitive than BA (P=.0.03)

Specificity and Positive Predictive values

Cultures	**SBS** False+ **Sp**(%)	**GRA** False+ **Sp**(%)	**SBID** False+ **Sp**(%)	**BA** False+ **Sp**(%)
Direct	41 (**89.5**)	0 (**100**)	22 (**94.3**)	45 (**88.4**)
After Lim	38 (**89.2**)	0 (**100**)	17 (**95.1**)	59 (**83.3**)
	Positive Predictive Value (%)			
Direct	**71.5**	**100**	**80.9**	**62.8**
After Lim	**77.9**	**100**	**87.9**	**65.7**

Description of False positive "GBS" characteristic colonies isolated on SBS and SBID

Agar	**Characteristic colonies not identified as GBS**
SBS	*S.pyogenes*, group C streptococci, group D streptococci, *Enterococcus sp.*, *S.bovis*, viridans streptococci, *Staphylococcus sp.*, yeasts, Gram negative bacilli*
SBID	Group C streptococci, *S.constellatus*, viridans streptococci, *Staphylococcus sp.*, Gram negative bacilli*

** Gram negative bacilli were very rarely detected and the colonies were of the respective expected colours but the colonies were not typical of GBS.*

DISCUSSION & CONCLUSION

➔**SBS**
- ➔**Very high sensitivity** for growth and detection of GBS.
- ➔The highest sensitivity (NS) by comparison to SBID and GRA.
- ➔More sensitive than BA in direct culture (p=0.03).

➔**SBS, GRA and SBID**
- ➔Reading highly improved after **48 h incubation.**
- ➔**GBS characteristic colonies easily detected** within a mixed flora even if GBS in low numbers.

➔**SBS as SBID**
- ➔**Aerobic condition** for incubation.
- ➔**Not 100 % specific**, confirmation needed for presumptive GBS.
- ➔**Detection of non-hemolytic** colonies.

➔**GRA**
- ➔**Anaerobic condition** for incubation.
- ➔**100% specific**, no need for GBS confirmation tests: **lowest workload.**
- ➔Non-hemolytic strains undetected.

➔**BA**
- ➔The **less sensitive** agar.
- ➔**Not 100 % specific**, confirmation needed for presumptive GBS.
- ➔Non-hemolytic strains undetected in mixed culture.

StreptoB Select, a new chromogenic medium, has demonstrated very good performances for GBS prenatal screening culture of vagino-rectal swabs.

To perform GBS prenatal screening cultures, the lowest workload with the highest sensitivity could be expected with a combination use of SBS or SBID with GRA for Lim sub-cultures.

REFERENCES

- Prevention of perinatal group B streptococcal diseases: Revised guidelines from CDC. MMWR 2002;51 (RR-11), 1-22
- Prevention of perinatal group B streptococcal infections: Belgian guidelines 2003, Belgian Superior Council of Hygiene, SCH 7721.

Acknowledgments to Biorad France, for the fund supporting the study.