PROCESSING SWABS FOR GROUP B STREPTOCOCCAL CARRIAGE

BSOP 58

Issued by Standards Unit, Evaluations and Standards Laboratory
Centre for Infections
STATUS OF NATIONAL STANDARD METHODS

National Standard Methods, which include standard operating procedures (SOPs), algorithms and guidance notes, promote high quality practices and help to assure the comparability of diagnostic information obtained in different laboratories. This in turn facilitates standardisation of surveillance underpinned by research, development and audit and promotes public health and patient confidence in their healthcare services. The methods are well referenced and represent a good minimum standard for clinical and public health microbiology. However, in using National Standard Methods, laboratories should take account of local requirements and may need to undertake additional investigations. The methods also provide a reference point for method development.

National Standard Methods are developed, reviewed and updated through an open and wide consultation process where the views of all participants are considered and the resulting documents reflect the majority agreement of contributors.

Representatives of several professional organisations, including those whose logos appear on the front cover, are members of the working groups which develop National Standard Methods. Inclusion of an organisation’s logo on the front cover implies support for the objectives and process of preparing standard methods. The representatives participate in the development of the National Standard Methods but their views are not necessarily those of the entire organisation of which they are a member. The current list of participating organisations can be obtained by emailing standards@hpa.org.uk.

The performance of standard methods depends on the quality of reagents, equipment, commercial and in-house test procedures. Laboratories should ensure that these have been validated and shown to be fit for purpose. Internal and external quality assurance procedures should also be in place.

Whereas every care has been taken in the preparation of this publication, the Health Protection Agency or any supporting organisation cannot be responsible for the accuracy of any statement or representation made or the consequences arising from the use of or alteration to any information contained in it. These procedures are intended solely as a general resource for practising professionals in the field, operating in the UK, and specialist advice should be obtained where necessary. If you make any changes to this publication, it must be made clear where changes have been made to the original document. The Health Protection Agency (HPA) should at all times be acknowledged.

The HPA is an independent organisation dedicated to protecting people’s health. It brings together the expertise formerly in a number of official organisations. More information about the HPA can be found at www.hpa.org.uk.

The HPA aims to be a fully Caldicott compliant organisation. It seeks to take every possible precaution to prevent unauthorised disclosure of patient details and to ensure that patient-related records are kept under secure conditions.

More details can be found on the website at www.evaluations-standards.org.uk. Contributions to the development of the documents can be made by contacting standards@hpa.org.uk.

Please note the references are now formatted using Reference Manager software. If you alter or delete text without Reference Manager installed on your computer, the references will not be updated automatically.

Citation for this document:
INDEX

STATUS OF NATIONAL STANDARD METHODS .............................................................. 2
INDEX .......................................................................................................................... 3
AMENDMENT PROCEDURE ....................................................................................... 4
SCOPE OF DOCUMENT ............................................................................................ 5
INTRODUCTION ........................................................................................................... 5
  LANCEFIELD GROUP B STREPTOCOCCI ............................................................... 5
  INFECTION .............................................................................................................. 5
  METHOD OF INVESTIGATION ................................................................................ 6
  TREATMENT ............................................................................................................ 6
1.0 SAFETY CONSIDERATIONS ................................................................................ 7
  1.1 SPECIMEN COLLECTION ................................................................................... 7
  1.2 SPECIMEN TRANSPORT AND STORAGE .......................................................... 7
  1.3 SPECIMEN PROCESSING ................................................................................... 7
2.0 SPECIMEN COLLECTION .................................................................................... 7
  2.1 OPTIMAL TIME OF SPECIMEN COLLECTION .................................................... 7
  2.2 CORRECT SPECIMEN TYPE AND METHOD OF COLLECTION ......................... 7
  2.3 ADEQUATE QUANTITY AND APPROPRIATE NUMBER OF SPECIMENS .......... 7
3.0 SPECIMEN TRANSPORT AND STORAGE .......................................................... 7
  3.1 TIME BETWEEN SPECIMEN COLLECTION AND PROCESSING ....................... 7
  3.2 SPECIAL CONSIDERATIONS TO MINIMISE DETERIORATION ......................... 7
4.0 SPECIMEN PROCESSING .................................................................................... 8
  4.1 TEST SELECTION ............................................................................................... 8
  4.2 APPEARANCE .................................................................................................... 8
  4.3 MICROSCOPY .................................................................................................... 8
  4.4 CULTURE AND INVESTIGATION ...................................................................... 8
  4.5 IDENTIFICATION ............................................................................................... 8
  4.6 ANTIBIOTIC SUSCEPTIBILITY TESTING ............................................................ 9
5.0 REPORTING PROCEDURE ................................................................................... 9
  5.1 MICROSCOPY .................................................................................................... 9
  5.2 CULTURE ........................................................................................................... 9
6.0 REPORTING TO THE HPA (LOCAL AND REGIONAL SERVICES AND CDSC CENTRE FOR INFECTIONS) ........................................................................... 9
ACKNOWLEDGEMENT AND CONTACTS ................................................................. 9
REFERENCES ........................................................................................................... 11
AMENDMENT PROCEDURE

<table>
<thead>
<tr>
<th>Amendment Number/Date</th>
<th>Issue no. Discarded</th>
<th>Insert Issue no.</th>
<th>Page</th>
<th>Section(s) involved</th>
<th>Amendment</th>
</tr>
</thead>
<tbody>
<tr>
<td>3/09/06/2006</td>
<td>2</td>
<td>2.1</td>
<td>1</td>
<td>Title</td>
<td>Title amended</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>5</td>
<td>Scope of Document</td>
<td>New reference added</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>6</td>
<td>Screening Method</td>
<td>Title of section renamed</td>
</tr>
</tbody>
</table>

Each National Standard Method has an individual record of amendments. The current amendments are listed on this page. The amendment history is available from standards@hpa.org.uk.

On issue of revised or new pages each controlled document should be updated by the copyholder in the laboratory.
PROCESSING SWABS FOR GROUP B STREPTOCOCCAL CARRIAGE

Type of specimens: Vaginal swabs
Rectal swabs

SCOPE OF DOCUMENT

This SOP describes the processing of specimens from pregnant women for carriage of Group B streptococci (GBS). Recognising that it is not recommended in the UK to screen routinely for GBS (Royal College of Obstetricians and Gynaecologists 2003; National Institute of Clinical Excellence 2003; HPA 2004), this SOP provides a standardised method for culture where clinicians decide to investigate specific patients with conditions considered to confer a high risk of infection. Commercial tests detecting the GBS group antigen extracted from vaginal swabs based upon latex agglutination, ELISA, immunofiltration, immunochromatography, optical immunoassay and other methods are available. However, the evidence accumulated has shown that the sensitivity and specificity of direct antigen tests is inferior to that of culture methods.

INTRODUCTION

LANCÉFIELD GROUP B STREPTOCOCCI

Lancefield Group B streptococci, or Streptococcus agalactiae, are oxidase-negative, catalase-negative Gram-positive cocci occurring in chains. GBS are facultative anaerobes that are serologically classified on the basis of cell wall polysaccharide antigens. On blood agar, the species exhibit \( \beta \)-haemolysis. This can be used as an early step in identifying clinical isolates. After 18-24 hours incubation at 35-37°C colonies tend to be slightly larger than other streptococci (approximately 1mm) and have a less distinct zone of \( \beta \)-haemolysis.

GBS normally colonises the vagina in many women and the intestines of men and women. Up to 30% of women carry GBS in the vagina or rectum without it causing problems or symptoms. The gastrointestinal tract is the likely human reservoir of GBS, with the genitourinary tract the most common site of secondary spread.

INFECTION

GBS may cause potentially devastating early onset disease primarily in newborns, pregnant women, and adults with underlying medical conditions (eg diabetes mellitus). In pregnancy this organism can infect the amniotic fluid (see BSOP 26 – Investigation of fluids from normally sterile sites), which can lead to neonatal sepsis, pneumonia and meningitis (see BSOP 23 – Investigation of gastric aspirates and infection screen swabs from neonates).

In pregnant women, GBS infection causes urinary tract infection, amnionitis, endometritis, wound infection; stillbirths and premature delivery have also been attributed to GBS. In non-pregnant adults, skin or soft tissue infection, bacteraemia, genitourinary infection, and pneumonia are the most common manifestations of disease.

Neonatal infection refers to infection occurring during the first four weeks of life. Infection may be superficial and localised (eg conjunctivitis, pustules, skin infection), deep and localised (pneumonia, septic arthritis) or systemic (sepsicaemia, meningitis). Presentation differs according to age at onset: early onset disease is more likely than late onset to present with sepsis.
In 1998, a Working Group was established by the then Public Health Laboratory Service (now the Health Protection Agency) with a remit to assess the burden of GBS disease and to produce evidence based national guidelines for the control and prevention of invasive neonatal GBS disease. As part of this programme, enhanced surveillance was undertaken in conjunction with the British Paediatric Surveillance Unit (London). The surveillance showed an incidence of 0.74 cases per 1000 live births and a mortality rate of 9.7%. The predominant GBS serotypes were III, Ia and V.

The incidence of infection also increases with low birth weight or prematurity and may be divided into:

- **early onset (0-6 days)** - this occurs in the first six days (usually within 48 hours) of life and is caused by infection ascending from the maternal genital tract or, less commonly, via the placenta. Only a small percentage of infants colonised with this organism develop early onset disease. Early infections tend to be associated with pneumonia and sepsicaemia and may be confused with respiratory distress syndrome.

- **late onset (7-90 days)** - this occurs after the first six days (7-90 days) and is associated with acquisition of the organism through vertical or nosocomial transmission or from the external (e.g., hospital) environment. GBS initially colonise the superficial sites and upper respiratory tract and progress to cause widespread sepsis. Late infection is more likely to be associated with meningitis.

### METHOD OF INVESTIGATION

In the UK, the advantages of screening pregnant women for colonisation with GBS routinely have not been demonstrated. However, according to local protocols, patients judged clinically to be at high risk for the development of Group B streptococcal infection may be investigated for carriage. The isolation rate of GBS from clinical specimens depends on several factors. Studies have shown that the accuracy of prenatal screening cultures for identification of GBS colonisation can be enhanced by attention to the timing of cultures, the sites swabbed and the microbiological method used for culture of organisms. Collection of swabs between 35 and 37 weeks gestation is recommended to improve the sensitivity and specificity of detection of colonisation at the time of delivery. Optimum yield will be achieved by selective/enrichment procedures applied to swabs obtained from the vagina and the anorectum, which increases the likelihood of GBS isolation by up to 30% compared with vaginal or cervical culture alone. Vaginal and rectal swabs are likely to isolate a diverse array of normal flora and use of selective enrichment broth is recommended to avoid overgrowth of other organisms.

### TREATMENT

Any neonate showing symptoms of early onset GBS disease should be treated with broad-spectrum antibiotics to cover GBS as well as other common pathogens, as neonatal sepsis can be rapidly fatal.

The treatment of pregnant women colonised with GBS is not recommended. However, intrapartum (i.e., during labour and delivery) antibiotics given to high-risk mothers with GBS may prevent ascending infection and subsequent early-onset streptococcal disease. The HPA Group B Streptococcus Working Group has produced interim guidelines for the prevention of early onset neonatal Group B streptococcal (GBS) infection in the UK. Further, the Royal College of Obstetricians and Gynaecologists has developed more detailed guidance in liaison with the HPA Working Group, for obstetricians, midwives and neonatologists on the prevention of early onset neonatal group B streptococcal infection.

In contrast to the UK, current USA guidelines advise that all women colonised with GBS at 35 to 37 weeks gestation should be offered intrapartum antibiotic prophylaxis in the form of high-dose penicillin or ampicillin.

---

**PROCESSING SWABS FOR GROUP B STREPTOCOCCAL CARRIAGE**

Issue no: 2 Issue date: 26.06.06 Issued by: Standards Unit, Evaluations and Standards Laboratory Page 6 of 12

Reference no: BSOP 582.1

This SOP should be used in conjunction with the series of other SOPs from the Health Protection Agency

www.evaluations-standards.org.uk

e-mail standards@hpa.org.uk
1.0 SAFETY CONSIDERATIONS

1.1 SPECIMEN COLLECTION

N/A

1.2 SPECIMEN TRANSPORT AND STORAGE

Swabs
Sealed plastic bag

1.3 SPECIMEN PROCESSING

Containment Level 2
All work which is likely to generate aerosols should be performed in a microbiological safety cabinet
The above guidance should be supplemented with local COSHH and risk assessments
Compliance with postal and transport regulations is essential

2.0 SPECIMEN COLLECTION

2.1 OPTIMAL TIME OF SPECIMEN COLLECTION

Before antimicrobial therapy where possible

2.2 CORRECT SPECIMEN TYPE AND METHOD OF COLLECTION

Swab the lower vagina (vaginal introitus) and the rectum with the same swab or two different swabs
Cervical swabs are not recommended
Swabs in Amies transport medium with charcoal

2.3 ADEQUATE QUANTITY AND APPROPRIATE NUMBER OF SPECIMENS

One combined vaginal/rectal swab or two separate swabs processed as one

3.0 SPECIMEN TRANSPORT AND STORAGE

3.1 TIME BETWEEN SPECIMEN COLLECTION AND PROCESSING

Specimens should be transported and processed as soon as possible

3.2 SPECIAL CONSIDERATIONS TO MINIMISE DETERIORATION

Swabs should be transported in Amies transport medium with charcoal
If processing is delayed, refrigeration is preferable to storage at ambient temperature. Delays of over 48h are undesirable
4.0 SPECIMEN PROCESSING

4.1 TEST SELECTION

N/A

4.2 APPEARANCE

N/A

4.3 MICROSCOPY

N/A

4.4 CULTURE AND INVESTIGATION

4.4.1 Pre-treatment

N/A

4.4.2 Specimen processing

Enrichment Culture

Remove the cap aseptically from the container and place the swab(s) in the broth, break off (or cut) the swab stick(s) and replace the cap. Caps should be kept loose during incubation.

After incubation, sub-culture with a sterile loop and inoculate appropriate media (see table 4.4.3).

For the isolation of individual colonies, spread inoculum with a sterile loop.

4.4.3 Culture media, conditions and organisms for all specimens:

<table>
<thead>
<tr>
<th>Clinical details/conditions</th>
<th>Standard media</th>
<th>Incubation</th>
<th>Cultures read</th>
<th>Target organism(s)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Enrichment Culture</td>
<td>LIM Broth (10mL Todd-Hewitt broth supplemented with 10µg/mL colistin and 15µg/mL nalidixic acid)</td>
<td>35-37</td>
<td>18-24h</td>
<td>Group B streptococci</td>
</tr>
<tr>
<td></td>
<td>Then subculture to blood agar</td>
<td>35-37</td>
<td>40-48h</td>
<td></td>
</tr>
</tbody>
</table>

†The bottle should contain a volume of broth sufficient to cover the swabs

4.5 IDENTIFICATION

4.5.1 Minimum level in the laboratory

*Streptococcus agalactiae* species level (See BSOP ID 4 – Identification of *Streptococcus* species, *Enterococcus* species and morphologically similar organisms)
4.5.2. **Referral to Reference Laboratories**

All isolates from sterile sites, organisms with unusual or unexpected resistance, and whenever there is a laboratory or clinical problem (e.g., outbreak) or anomaly that requires elucidation

4.6 **ANTIBIOTIC SUSCEPTIBILITY TESTING**

Refer to SOP on Susceptibility Testing (BSOP 45)

5.0 **REPORTING PROCEDURE**

5.1 **MICROSCOPY**

N/A

5.2 **CULTURE**

Report:

*Negatives*

“Group B streptococci not isolated”

*Positives*

“Group B streptococci isolated”

5.2.1. **Culture reporting time**

Clinically urgent results: to be telephoned or sent electronically

Written report: 16 - 72h stating, if appropriate, that a further report will be issued

5.2.2. **Antibiotic Susceptibility testing**

Report susceptibilities as clinically indicated

6.0 **REPORTING TO THE HPA (LOCAL AND REGIONAL SERVICES AND CDSC CENTRE FOR INFECTIONS)**

Refer to the following:

Individual SOPs on organism identification

Health Protection Agency publications:

"Reporting to the CDR: A guide for laboratories"

"Hospital infection control: Guidance on the control of infection in hospitals"

Refer to current guidelines on CDSC and COSURV reporting

Local guidelines

**ACKNOWLEDGEMENT AND CONTACTS**

This Standard Method was initiated and developed by the Group B Streptococcus Working Group for the HPA in conjunction with the Standard Methods Working Group for Bacteriology (http://www.hpa-standardmethods.org.uk/wg_bacteriology.asp). The contributions of many individuals in clinical bacteriology laboratories and specialist organisations who have provided information and comment during the development of this document, and final editing by the Medical Editor are also acknowledged.
The National Standard Methods are issued by Standards Unit, Evaluations and Standards Laboratory, Centre for Infections, Health Protection Agency London.

For further information please contact us at:

Standards Unit
Evaluations and Standards Laboratory
Centre for Infections
Health Protection Agency
Colindale, London
NW9 5EQ

e-mail standards@hpa.org.uk
REFERENCES


