

NATIONAL STANDARD METHOD

INVESTIGATION OF EYE SWABS AND CANALICULAR PUS

BSOP 2

Issued by Standards Unit, Evaluations and Standards Laboratory
Centre for Infections



Association of Medical Microbiologists
Association of Medical Microbiologists



INVESTIGATION OF EYE SWABS AND CANALICULAR PUS

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AMENDMENT PROCEDURE

Controlled document reference	BSOP 2
Controlled document title	Investigation of eye swabs and canalicular pus

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.

Amendment Number/ Date	Issue no. Discarded	Insert Issue no.	Page	Section(s) Involved	Amendment
6/ 05.03.08	4.1	5	All	All	PDF links inserted to cross-reference NSM documents Northern Ireland logo added Links to reference laboratory user manuals inserted. References reviewed and updated
			1	Front page	
			11	4.5.2 Referrals	
			14	References	

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INVESTIGATION OF EYE SWABS AND CANALICULAR PUS

Type of specimen: Eye swabs
Canalicular pus

SCOPE OF DOCUMENT

This National Standard Method (NSM) describes the processing and bacteriological investigation of specimens from the eyes with the exception of those from keratitis, endophthalmitis, hypopyon and post surgical infections for these refer to [BSOP 52 - Investigation of intraocular fluids and corneal scrapings](#). New molecular techniques are now available to diagnose chlamydia infections from eye swabs. These are not covered in this NSM.

INTRODUCTION

Infections of the eye can be caused by a variety of microorganisms. Swabs from eyes may be contaminated with skin microflora, but any organism may be considered for further investigation if clinically indicated.

Exogenous organisms may be introduced to the eye via hands, fomites (eg contact lenses), traumatic injury involving a foreign body², following surgery³, or simply by spread from adjacent sites.

Infections

Common mild eye infections include conjunctivitis (inflammation of the conjunctiva) and blepharitis (inflammation of the eyelid). Conjunctivitis may occur in association with infection of the eyelid (blepharoconjunctivitis) or of the cornea (keratoconjunctivitis). Less common and more severe infections include keratitis (inflammation of the cornea) and endophthalmitis (infection inside the eye itself). Haematogenous spread from a focus elsewhere in the body can also occur⁴. Other periocular infections include dacryoadenitis (inflammation of the lacrimal gland), dacryocystitis (inflammation of the lacrimal sac)⁵, canaliculitis (infection of the lacrimal puncta and canaliculi), and preseptal and orbital cellulitis. Invasive specimens may be required for optimal investigation of severe eye infections, and these are dealt with in [BSOP 52 - Investigation of intraocular fluids and corneal scrapings](#). Separate swabs in appropriate transport media are needed for the diagnosis of viral and chlamydial infections.

Eye infections occurring in the first four weeks of life caused by *Chlamydia trachomatis* or *Neisseria gonorrhoeae* are notifiable as ophthalmia neonatorum.

Eye swabs may be received from patients with any of these conditions but may need handling differently according to the type and severity of infection.

Blepharitis is associated with^{6,7}:

- *Staphylococcus aureus*
- *Staphylococcus epidermidis*
- *Corynebacterium* species
- *Propionibacterium acnes*

However, these organisms may be isolated from the eyelids of normal healthy individuals, necessitating careful interpretation of such cultures.

Conjunctivitis may be acute or chronic. The conjunctiva is the most commonly infected ocular tissue, and infectious conjunctivitis is one of the most common causes of red or sticky eyes. Common bacterial causes include:

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- *S. aureus*
- *Streptococcus pneumoniae*
- *Haemophilus influenzae*

Less common causes include Lancefield group A, C and G streptococci⁸, *Neisseria cinerea*⁹, *P. acnes*, *Moraxella* species and other Gram-negative rods, and anaerobes such as *Eubacterium* species and *Peptostreptococcus* species^{10,11}. *Moraxella catarrhalis* causes acute conjunctivitis and *Moraxella lacunata* causes a chronic infection¹¹. However, many of these organisms may also be isolated from the surrounding areas (skin), and so the interpretation of the significance of their presence is difficult.

Conjunctivitis caused by *Neisseria* species is uncommon in developed countries. The most important ocular pathogen in this genus is *Neisseria gonorrhoeae*. In adults it is associated with concomitant genital infection. In neonates it is an important cause of ophthalmia neonatorum, which may cause blindness if left untreated. *Neisseria meningitidis* has also been implicated in hyperacute conjunctivitis. Treatment of this is important to reduce the risk of dissemination, and rifampicin prophylaxis may be indicated on household contacts and the patient to eliminate throat carriage.

Conjunctivitis in neonates is caused by the pathogens commonly found in adult cases^{10,12}. Additional organisms include¹¹:

- *N. gonorrhoeae*
- *Haemophilus parainfluenzae*
- Lancefield group B streptococci and enterococci
- Enterobacteriaceae eg *Klebsiella pneumoniae* and *Proteus mirabilis*
- *Pseudomonas aeruginosa*

Chlamydial and viral conjunctivitis also occur. Inclusion conjunctivitis and trachoma are caused by various serotypes of *Chlamydia trachomatis*. Trachoma is associated with serotypes A-C. This occurs in rural under-developed areas, whereas inclusion conjunctivitis is associated with types D-K, and is a feature of developed urban communities¹³. These serotypes are associated with sexual transmission. The most common causes of viral conjunctivitis are adenoviruses.

***Acanthamoeba* species** can cause severe keratitis, usually in contact lens wearers or after ocular trauma. These protozoa may be isolated from corneal scrapings, as well as from contact lenses and storage cases ([BSOP 52 - Investigation of intraocular fluids and corneal scrapings](#) and [BSOP 31 - Investigation of specimens other than blood for parasites](#)).

Orbital cellulitis is the infection of orbital tissue. It can result from trauma, surgery, or an extension of paranasal sinus infections. It is a serious infection and may cause blindness, septic thrombosis of the cavernous sinus or intracranial infections. The most common pathogens in adults are *S. aureus*, streptococci and anaerobes. In children *H. influenzae* still remains prevalent, but the capsulated (type b) strain is rarely seen. Streptococci, staphylococci, peptostreptococci and *P. aeruginosa* may cause necrosis¹⁴. Eye swabs are of limited value in the investigation of orbital and preseptal cellulitis. Ideally aspirates from the affected tissues should be obtained and treated according to the procedures outlined in [BSOP 26 - Investigation of fluids from normally sterile sites](#). Blood cultures are also useful in diagnosis (see [BSOP 37 - Investigation of blood cultures \(for organisms other than Mycobacterium species\)](#)).

Canaliculitis is a rare condition. Infections are usually chronic and caused by anaerobic actinomycetes such as *Actinomyces israelii* or by *Propionibacterium propionicus*^{15,16}. Swabs of samples of the canalicular pus are preferable to eye swabs for diagnosis.

For further information about serious eye infections, including examination for *Acanthamoeba* species, refer to [BSOP 52 - Investigation of intraocular fluids and corneal scrapings](#).

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TECHNICAL INFORMATION/LIMITATIONS

Superficial swabs, although not ideal, may be all that is available. Deep-seated samples if available should be sought.

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1 SAFETY CONSIDERATIONS¹⁷⁻²⁷

1.1 SPECIMEN COLLECTION

N/A

1.2 SPECIMEN TRANSPORT AND STORAGE

Swabs

Sealed plastic bag.

Canalicular pus

Sterile leakproof container in a sealed plastic bag.

1.3 SPECIMEN PROCESSING

Containment Level 2 unless infection with a Hazard Group 3 organism, for example *M. tuberculosis*, brucella, or agent of exotic imported mycosis is suspected, in which case all work must be undertaken in a microbiological safety cabinet at Containment Level 3.

Refer to current guidance on the safe handling of all organisms documented in this NSM.

The above guidance should be supplemented with local COSHH and risk assessments. Compliance with postal and transport regulations is essential

2 SPECIMEN COLLECTION

2.1 OPTIMAL TIME OF SPECIMEN COLLECTION

Before antimicrobial therapy where possible, and preferably before application of local anaesthetic.

2.2 CORRECT SPECIMEN TYPE AND METHOD OF COLLECTION

Any available pus should be sampled as well as the lesion of interest.

Separate samples must be collected into appropriate transport media for detection of viruses or chlamydiae. Alcohol or acetone fixed smears for immunofluorescence are also used for chlamydial investigations.

For Acanthamoeba investigation see [BSOP 52 - Investigation of intraocular fluids and corneal scrapings](#) .

2.3 ADEQUATE QUANTITY AND APPROPRIATE NUMBER OF SPECIMENS

N/A

3 SPECIMEN TRANSPORT AND STORAGE

3.1 TIME BETWEEN SPECIMEN COLLECTION AND PROCESSING

Specimens should be transported and processed as soon as possible.

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3.2 SPECIAL CONSIDERATIONS TO MINIMISE DETERIORATION

Swabs should be transported in Amies transport medium with charcoal²⁸.

If processing is delayed, refrigeration is preferable to storage at ambient temperature. Delays of over 48h are undesirable.

4 SPECIMEN PROCESSING

4.1 TEST SELECTION

N/A

4.2 APPEARANCE

N/A

4.3 MICROSCOPY ([BSOFTP 39 – Staining Procedures](#))

Gram's stain

Eye swabs (from neonates with sticky eyes and others as appropriate) and canalicular pus

Prepare a thin smear from the swab or pus on a clean microscope slide for Gram's staining.

4.4 CULTURE AND INVESTIGATION

4.4.1 PRE-TREATMENT

N/A

4.4.2 SPECIMEN PROCESSING

Inoculate each agar plate with swab or pus ([QSOP 52 - Inoculation of culture media \(formerly BSOP 54\)](#)).

For inoculation methods performed at the patient's side, refer to local protocols.

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4.4.3 CULTURE MEDIA, CONDITIONS AND ORGANISMS FOR ALL SPECIMENS:

Clinical details/ conditions	Standard media	Incubation			Cultures read	Target organism(s)
		Temp °C	Atmos	Time		
Blepharitis Conjunctivitis Sticky eye If no clinical details available, treat as a 'sticky eye'	Chocolate agar	35-37	5-10% CO ₂	40-48 h	daily	<i>H. influenzae</i> Lancefield group A,B,C and G streptococci <i>Moraxella</i> species <i>N. gonorrhoeae</i> <i>N. meningitidis</i> <i>P. aeruginosa</i> <i>S. aureus</i> <i>S. pneumoniae</i> Other organisms (see section 4.5.1)
	Blood agar	35-37	5-10% CO ₂	40-48 h	daily	

For these situations, add the following:

Clinical details/ conditions	Supplementary media	Incubation			Cultures read	Target organism(s)
		Temp °C	Atmos	Time		
GUM clinic sticky eye Neonates	GC selective agar	35-37	5-10% CO ₂	40-48 h	≥40 h	<i>N. gonorrhoeae</i>
Immunocompromised Chronic blepharitis	Sabouraud agar	28-30	air	40-48 h*	≥40 h	Fungi
Canaliculitis † Orbital cellulitis Dacryocystitis † Dacryoadenitis † Keratitis‡ Endophthalmitis ‡ Hypopyon ‡ Post surgery ‡ Post trauma	Fastidious anaerobe agar	35-37	anaerobic	40-48 h*	≥40 h	Anaerobes
				10 d	≥40 h, at 7 d and 10 d	
	Sabouraud agar	28-30	air	40-48 h*	≥40 h	Fungi
If Gram-negative rods seen in Gram film	CLED agar	35-37	air	16-24 h	≥16 h	Enterobacteriaceae

Other organisms for consideration - *Chlamydia* species and viruses

*incubation may be extended to 5 days; in such cases plates should be read at ≥40 h and then left in the incubator/cabinet until day 5

†extend incubation time to 10 days if clinically suspected or Gram-positive branching rods present in Gram's stain

‡Refer to [BSOP 52 - Investigation of intraocular fluids and corneal scrapings](#)

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4.5 IDENTIFICATION

4.5.1 MINIMUM LEVEL IN THE LABORATORY

Actinomycetes	"actinomycetes" level
Anaerobes	"anaerobes" level BSOPID 14 - Identification of non-sporing, non-branching anaerobes BSOPID 8 - Identification of Clostridium species BSOPID 25 - Identification of anaerobic Gram-negative rods
Coagulase-negative staphylococci	"coagulase-negative" level
Diphtheroids	"diphtheroid" level
Enterobacteriaceae	"coliforms" level
Enterococci	species level
Fungi	genus level
Haemophilus influenzae	species level
Lancefield groups A, B, C and G streptococci	Lancefield group level
Moraxella species	species level
Neisseria meningitidis	species level
P. aeruginosa	species level
Pseudomonads	"pseudomonads" level
S. aureus	species level
S. pneumoniae	species level
α-haemolytic streptococci	"α-haemolytic" level
Yeasts	"yeasts" level

Organisms may be further identified if clinically or epidemiologically indicated

4.5.2 REFERRAL TO REFERENCE LABORATORIES

For information on the tests offered, turn around times, transport procedure and the other requirements of the reference laboratory refer to the appropriate users manual as found by using the links below.

N. meningitidis for strain characterisation and antimicrobial susceptibility testing

http://www.hpa.org.uk/cfi/other_ref_labs/mru1.htm

Actinomycetes for strain characterisation and antimicrobial susceptibility testing

<http://www.hpa.org.uk/cfi/ar/default.htm>

Fungi requiring identification and/or susceptibility testing

<http://www.hpa.org.uk/cfi/mycology/default.htm>

Organisms with unusual or unexpected resistance, and whenever there is a laboratory or clinical problem, or anomaly that requires elucidation should be sent to the appropriate reference laboratory.

4.6 ANTIMICROBIAL SUSCEPTIBILITY TESTING

Refer to NSM [BSOP 45 - Susceptibility Testing](#)

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5 REPORTING PROCEDURE

5.1 MICROSCOPY

Report on WBCs and organisms detected.

5.2 MICROSCOPY REPORTING TIME

Urgent microscopy results to be telephoned or sent electronically

Written report 16 – 72 h

5.3 CULTURE

Report:

Clinically significant organisms isolated or other growth, eg “No significant growth” or absence of growth

5.3.1 CULTURE REPORTING TIME

Clinically urgent results: to be telephoned or sent electronically.

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

5.3.2 ANTIBIOTIC SUSCEPTIBILITY TESTING

Report susceptibilities as clinically indicated

6 REPORTING TO THE HPA²⁹ (LOCAL AND REGIONAL SERVICES AND CENTRE FOR INFECTIONS)

Refer to the following:

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

Isolation of *N. meningitidis* should be reported to the CCDC

Ophthalmia neonatorum is a notifiable disease

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7 ACKNOWLEDGEMENTS AND CONTACTS

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