

NATIONAL STANDARD METHOD

# IDENTIFICATION OF *SHIGELLA* SPECIES

BSOP ID 20

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



Association of Medical Microbiologists  
*Association of Medical Microbiologists*  
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## **Suggested citation for this document:**

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# AMENDMENT PROCEDURE

<b>Controlled document reference</b>	<b>BSOP ID 20</b>
<b>Controlled document title</b>	<b>Identification of <i>Shigella</i> species</b>

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](mailto: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
2/ 09.11.07	1.1	2	1	<b>Front Page</b>	Northern Ireland logo added
			8	<b>Flow chart</b>	Title changed and flowchart put in to Visio format. Contents of flow chart updated.
			9	<b>6 Referrals</b>	Links to reference laboratory user manuals inserted.
			11	<b>References</b>	References reviewed and updated
			All	<b>All</b>	PDF links inserted to cross-reference NSM documents

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# IDENTIFICATION OF *SHIGELLA* SPECIES

## SCOPE OF DOCUMENT

This National Standard Method (NSM) describes the identification of *Shigella* species with particular reference to isolation from faeces.

## INTRODUCTION

### Taxonomy

The genus *Shigella* belongs to the family Enterobacteriaceae and consists of four species; *Shigella dysenteriae*, *Shigella flexneri*, *Shigella boydii*, and *Shigella sonnei*. Each of the species, with the exception of *S. sonnei*, is subdivided by serotype.

### Characteristics

*Shigella* species are small Gram-negative rods. They produce pink colonies on XLD medium and colourless colonies on DCA. *Shigella* species are facultative anaerobes, are non-motile, oxidase-negative, urease-negative, do not decarboxylate lysine and all except *S. dysenteriae* type 1 are catalase-positive<sup>2</sup>. The species may be differentiated by biochemical tests and serology of their lipopolysaccharides<sup>3</sup>. The majority of *Shigella* species, except *S. flexneri* 6, and *S. boydii* 13 and 14, ferment sugars without gas production. *S. boydii*, *S. flexneri* and *S. sonnei*, with a few exceptions, ferment mannitol; *S. dysenteriae* does not. *S. sonnei*, and *S. dysenteriae* type 1 are the only species that are ONPG-positive. *S. boydii* 13 are Ornithine positive, and some may be ONPG positive.

*Shigella* species are highly infective. The infective dose is particularly low with *S. dysenteriae*, which may require as few as 10-100 organisms to cause infection<sup>3</sup>.

### Principles of identification

Isolates from primary culture are identified by colonial appearance, biochemical tests and serology (agglutination with specific antisera). *Plesiomonas shigelloides* cross reacts with *S. sonnei* antisera. If confirmation of identification is required, isolates should be sent to the Reference Laboratory. All identification tests should ideally be performed from non-selective agar.

## TECHNICAL INFORMATION

N/A

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# 1 SAFETY CONSIDERATIONS<sup>4-14</sup>

Most *Shigella* species are in Hazard Group 2. An important exception is *Shigella dysenteriae* type 1. All work on *Shigella dysenteriae* type 1 must be performed under Containment level 3 conditions.

*Shigella dysenteriae* type 1 causes severe and sometimes fatal disease.

Laboratory acquired infections have been reported. Low numbers of *Shigella* species are required for an infective dose<sup>3</sup>.

Refer to current guidance on the safe handling of all Hazard Group 2 organisms documented in this NSM.

Laboratory procedures that give rise to infectious aerosols must be conducted 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 TARGET ORGANISMS<sup>15</sup>

## Genus *Shigella*

All species cause human infections

*Shigella dysenteriae* (15 serotypes)

*Shigella boydii* (20 serotypes)

*Shigella flexneri* (6 serotypes which can be sub-divided into sub-serotypes)

*Shigella sonnei* (1 serotype, 2 variants - rough and smooth)

# 3 IDENTIFICATION

## 3.1 MICROSCOPIC APPEARANCE

N/A

## 3.2 PRIMARY ISOLATION MEDIA

XLD agar incubated in air at 35 - 37°C for 18 – 24 h

DCA incubated in air at 35 - 37°C for 18 – 24 h

## 3.3 COLONIAL APPEARANCE

*Shigella* species on XLD agar produce 1 - 2 mm diameter red colonies (no black centre). Colonies on DCA are colourless (*S. sonnei* may form pale pink colonies because of late lactose fermentation).

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### **3.4 TEST PROCEDURES**

#### **3.4.1 AGGLUTINATION**

Agglutination with shigella antiserum (not all the serotypes are contained in polyvalent antisera).

#### **3.4.2 BIOCHEMICAL TESTS**

Urease (see [BSOFTP 36 - Urease Test](#))

*Shigella* species do not produce urease

Oxidase (optional) (see [BSOFTP 26 - Oxidase Test](#))

*Shigella* species are oxidase-negative

Commercial identification kit

In house identification kit

### **3.5 FURTHER IDENTIFICATION**

N/A

### **3.6 STORAGE AND REFERRAL**

If required, save the pure isolate on a nutrient agar slope for referral to the Reference Laboratory.

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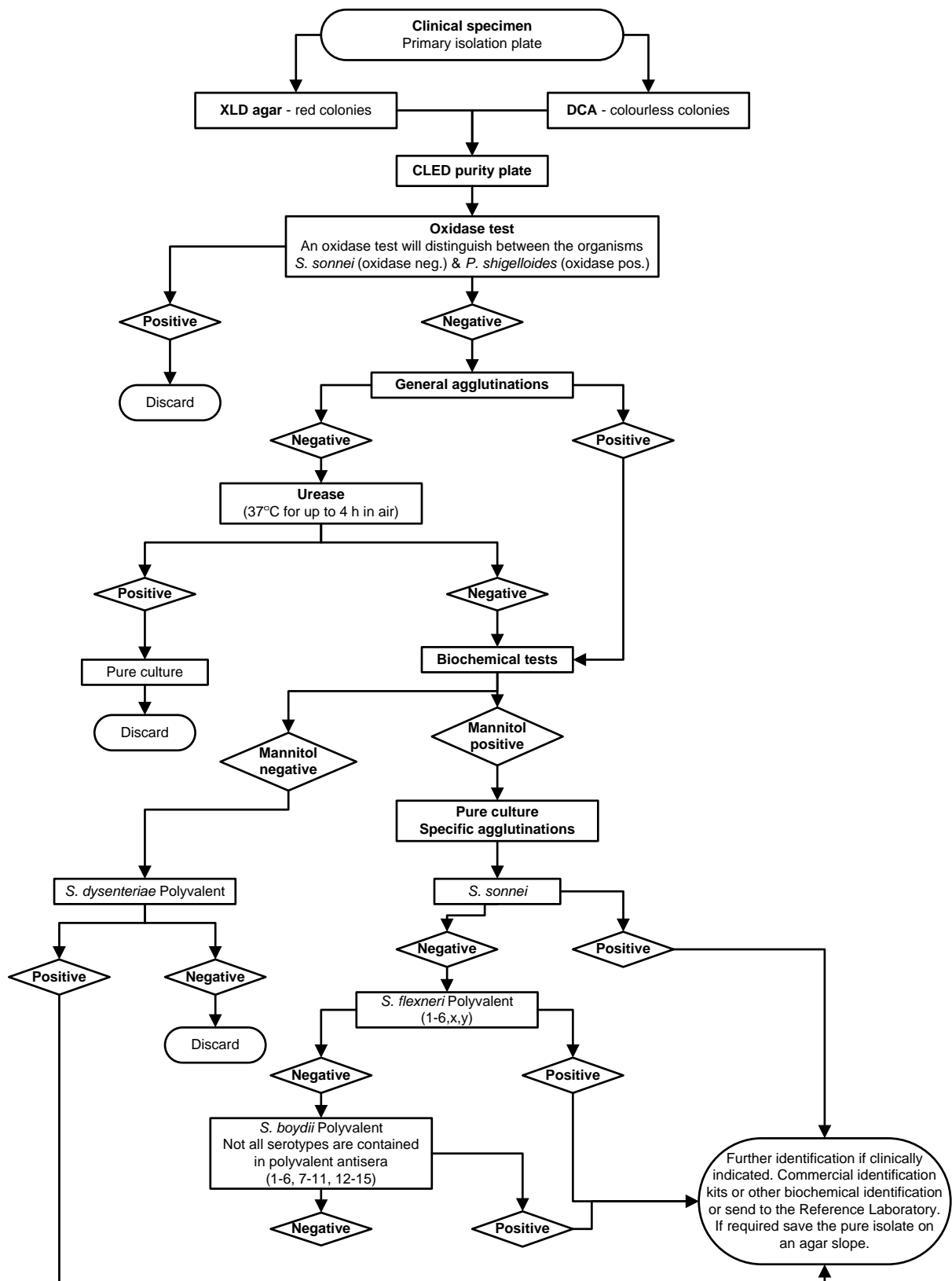
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## 4 IDENTIFICATION OF *SHIGELLA* – FLOW CHART



The flow chart is for guidance only

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## 5 REPORTING

### 5.1. PRESUMPTIVE IDENTIFICATION

If appropriate growth characteristics, colonial appearance, urease and serology results are demonstrated.

### 5.2 CONFIRMATION OF IDENTIFICATION

Following use of commercial or in-house identification kit results and/or the Reference Laboratory report.

### 5.3 MEDICAL MICROBIOLOGIST

Inform the medical microbiologist of presumptive or confirmed *Shigella dysenteriae* O1 isolates, according to local protocols.

The medical microbiologist should also be informed of a presumptive or confirmed *Shigella* species if the request card bears relevant information eg

- enterocolitis, dysentery (especially if complicated by haemolytic-uraemic syndrome)
- neurological dysfunction or confusional states
- history of recent foreign travel or laboratory work
- food poisoning
- investigations of outbreak situations

Follow local protocols for reporting to clinician

### 5.4 CCDC

Refer to local Memorandum of Understanding.

### 5.5 CENTRE FOR INFECTIONS<sup>16</sup>

Refer to current guidelines on CDSC and COSURV reporting.

### 5.6 INFECTION CONTROL STAFF

Inform the infection control team of presumptive or confirmed isolates of *Shigella* species.

## 6 REFERRALS

### 6.1 REFERENCE LABORATORY

For information on the tests offered, turn around times, transport procedure and the other requirements of the reference laboratory refer to: <http://www.hpa.org.uk/cfi/lep/default.htm>

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## 7 ACKNOWLEDGEMENTS AND CONTACTS

This National Standard Method has been developed, reviewed and revised by the National Standard Methods Working Group for Clinical Bacteriology ([http://www.hpa-standardmethods.org.uk/wg\\_bacteriology.asp](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 acknowledged.

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