

SEROTYPING OF SHIGELLA

2012/04

1. INTENDED USE

Shigella antisera are designed for the serological identification of cultures of *Shigella* by the slide agglutination method, for epidemiological and diagnostic purposes.

These sera must be only used to study immobile strains of *Enterobacteria* presenting biochemical characters of *Shigella*.

2. PRINCIPE

The test is based on agglutination, by specific antisera, of bacteria possessing the corresponding antigens.

1) POLYVALENT SERA

Each polyvalent serum contains a number of serotypes of the 4 subgroups:

- **Subgroup A: 2 polyvalent sera**

This subgroup comprises 12 mannitol-negative serotypes.

- Serum A1: *Shigella dysenteriae* indole-negative: 1, 3, 4, 5, 6 antiserum.
- Serum A2: *Shigella dysenteriae* indole-positive: 2, 7, 8 antiserum.

Serotypes 9, 10, 11 and 12 are rare and the corresponding sera are therefore not included in these polyvalent sera.

- **Subgroup B: 1 polyvalent serum**

- *Shigella flexneri* antiserum agglutinates the 6 serotypes of this subgroup, as well as X and Y.

- **Subgroup C: 3 polyvalent sera**

- Serum C1: *Shigella boydii* indole-negative: 1, 2, 3, 4 antiserum.
- Serum C2: *Shigella boydii* indole-negative: 8, 10, 14 antiserum.
- Serum C3: *Shigella boydii* indole-positive: 5, 7, 9, 11, 15 antiserum.

Serotypes 12, 13, 16, 17 and 18 are rare and the corresponding sera are therefore not included in these polyvalent sera.

Serotype 6, closely related to *Shigella sonnei* phase II is agglutinated by the *Shigella sonnei* antiserum.

- **Subgroup D: 1 mixed serum**

- *Shigella sonnei* antiserum agglutinates the 2 phases of this species.

2) MONOVALENT SERUM

- Serum A1: *Shigella dysenteriae* 1 (mannitol -, indole -, catalase +) antiserum.

These antisera are obtained by immunization of rabbits with selected strains of *Shigella*.

3. HOW SUPPLIED

Polyvalent sera and the monovalent serum are presented in 2 ml dropper bottles (40 tests):

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| • Antiserum <i>Shigella flexneri</i> polyvalent | code 57151 |
| • Antiserum <i>Shigella sonnei</i> polyvalent | code 57171 |
| • Antiserum <i>Shigella dysenteriae</i> monovalent | code 57161 |
| • Antiserum <i>Shigella dysenteriae</i> polyvalent A1 + A2 | code 57182 |
| • Antiserum <i>Shigella boydii</i> polyvalent C1 + C2 + C3 | code 57183 |

4. STORAGE

Sera stored at +2-8°C in the absence of contamination are stable until the expiry date indicated on the kit (even when opened).

5. MATERIAL REQUIRED BUT NOT SUPPLIED

- Glass slide.
- Plastic or platinum inoculation loop.
- Physiological saline.

6. PRECAUTIONS FOR USE

- Always comply with current techniques and precautions concerning protection against microbiological hazards, for handling and elimination of material and biological products used for the agglutination reaction.
- These sera contain < 0.1% sodium azide. Sodium azide can react with lead or copper present in the plumbing forming explosive metallic azides. When eliminating these reagents, rinse abundantly with water to avoid the formation of azide deposits.
- Do not dilute reagents.

7. PROCEDURE

Serotyping is performed, after identification of the species, on a pure, fresh culture of *Shigella* isolated on non-selective agar medium.

Perform a control test on the strain to be tested in physiological saline:

- Take one loop of culture. **Only use plastic or platinum loops.**
- Suspend these bacteria in a drop of physiological saline, ensuring homogeneous suspension. No agglutination should be observed with physiological saline. If agglutination is observed, it corresponds to a self-agglutinating strain and the test with antisera cannot be performed.
- Deposit 1 drop of immune serum on the slide.
- Take one loop of fresh, pure culture of *Shigella*. **Only use plastic or platinum loops.**
- Suspend these bacteria in each drop of serum, ensuring homogeneous suspension gradually adding bacteria to the serum.
- Shake the slide with a gently rotary movement.
- Examine the mixture with the naked eye over a dark surface or over a concave mirror.

8. INTERPRETATION OF THE RESULTS

A positive reaction corresponds to the appearance of agglutination in a **maximum of one minute**.

9. TEST PERFORMANCES/QUALITY CONTROL

The activity of *Shigella* antisera is controlled with the following reference strains:

Serum	Strain tested
Antiserum <i>Shigella flexneri</i> polyvalent (57151)	<i>Shigella flexneri</i> 2 (NCDC 2747-71) <i>Shigella flexneri</i> 3 (NCDC 2783-71) <i>Shigella flexneri</i> 6 (NCDC 2924-71)
Antiserum <i>Shigella sonnei</i> polyvalent (57171)	<i>Shigella sonnei</i> Phase 1 (97-10968) <i>Shigella sonnei</i> Phase 2 (262/80) <i>Shigella sonnei</i> Mixte (248/81)
Antiserum <i>Shigella dysenteriae</i> monovalent (57161)	<i>Shigella dysenteriae</i> 1 (NCDC 1007-71)
Antiserum <i>Shigella dysenteriae</i> polyvalent A1 (57182)	<i>Shigella dysenteriae</i> 1 (NCDC 1007-71) <i>Shigella dysenteriae</i> 3 (NCDC 3690-73)
Antiserum <i>Shigella dysenteriae</i> polyvalent A2 (57182)	<i>Shigella dysenteriae</i> 2 (NCDC 4106-65) <i>Shigella dysenteriae</i> 7 (NCDC 4788-55)
Antiserum <i>Shigella boydii</i> polyvalent C1 (57183)	<i>Shigella boydii</i> 1 (NCDC 6310-65) <i>Shigella boydii</i> 3 (NCDC 67)
Antiserum <i>Shigella boydii</i> polyvalent C2 (57183)	<i>Shigella boydii</i> 8 (NCDC 3073-50) <i>Shigella boydii</i> 10 (NCDC 04-74)
Antiserum <i>Shigella boydii</i> polyvalent C3 (57183)	<i>Shigella boydii</i> 5 (NCDC 5393-79) <i>Shigella boydii</i> 11 (NCDC 3357-71)

10. MANUFACTURER'S QUALITY CONTROL

All reagents manufactured are prepared according to our Quality System, starting from reception of raw materials to final commercialization of the product. Each lot is submitted to quality control tests and is only released onto the market when it complies with predefined acceptance criteria. Records relating to production and control of each lot are kept by Bio-Rad.

11. LIMITS OF USE

Identification of the bacterial species must be performed before determination of the serogroup.

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