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MAST® ASSURE ANTISERUM SHIGELLA

Liquid stable antisera for the determination of O antigen types and groups for the serological identification of *Shigella.*

FOR IN VITRO DIAGNOSTIC USE ONLY

Contents: See pack label.

Formulation
MAST® ASSURE ANTISERUM are prepared from rabbits hyperimmunised with standard strains of killed organisms possessing known serotypes or group specific antigens and contain 0.085% sodium azide as preservative.
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Stability and storage
Store unopened at 2 to 8°C until the expiry date shown on the pack label. Once opened, MAST® ASSURE ANTISERUM should be stored at 2 to 8°C and may be used until the expiry date given on the label. **Do not freeze reagents.**

Warnings and precautions
For *in vitro* diagnostic use only. Observe approved biohazard precautions and aseptic techniques. To be used only by adequately trained and qualified laboratory personnel. Sterilise all biohazard waste before disposal. Sodium azide preservative may be toxic if ingested and may react with lead and copper plumbing to form highly explosive salts. Always dispose of by flushing to drain with plenty of water. Refer to Product Safety Data sheet.

Materials required but not provided
Standard microbiological supplies and equipment such as loops, applicator sticks, clean glass microscope slides or glass test tubes swabs, MAST culture media, incinerators and incubators, etc., as well as reagents and additives such as sterile 0.85% saline solution.

Procedure
Slide agglutination of live organisms
1. Dispense two 5 to 10 µl volumes of sterile 0.85% saline solution (saline) onto a carefully cleaned microscope slide. The slide may be partitioned using a chinagraph pencil. With a platinum wire or disposable inoculation loop take one 1 to 2mm colony of live organisms from a fresh culture on MAST® Nutrient Agar DM179 or similar and emulsify into each drop of saline to produce a distinct and uniform turbidity.
2. Place a drop (30 to 40 µl) of antiserum onto one of the emulsified isolates and on to the other a drop (30-40 µl) of saline as a control.
   Note: Do not allow the organism to contaminate the antiserum dropper bottle.
3. Mix the reagents by tilting the slide back and forth for 60 seconds while viewing it under indirect light against a dark background.
4. Distinct clumping or agglutination within this period, without clumping in the saline control (auto-agglutination), should be regarded as a positive result. Weak agglutination should be recorded as negative.

Slide agglutination of heat treated organisms
If the live cells do not produce positive agglutination, this is probably because some strains possess heat labile capsular (K) antigens which mask the presence of the heat stable somatic (O) antigens. Should this occur, prepare a dense cell suspension of the organism in saline and heat it to 100°C for 60 minutes or autoclave at 121°C for 15 minutes. Centrifuge at 900g for 20 minutes. Discard the supernatant and resuspend the pellet in saline to form a dense, homogeneous suspension. Repeat the slide agglutination tests as described above.

Interpretation of results
Isolates producing a distinct positive reaction with a polyvalent antiserum are assumed to be a *Shigella* from the Group (A-D) represented by the antiserum. Further testing of the isolate should be conducted as described in steps 1 to 3, with monovalent antisera. If the organism is identified as *Sh. flexneri* (Group B) it should be typed and grouped separately.

Limitations of use
Only cultures of organisms identified as *Shigella* by morphological and biochemical features should be serotyped with this product.
Polyvalent antisera are intended for use in rapid slide agglutination tests only. Monovalent antisera are intended for use in rapid slide agglutination tests for further identification.
Positive results may be confirmed by tube agglutination tests.

Quality control
It is recommended that quality control should be performed with at least one organism to demonstrate a positive reaction and at least one organism to demonstrate a negative reaction. Do not use the product if the reactions with the control organisms are incorrect. Check for signs of deterioration. Do not use reagents if they are contaminated or cloudy.

References
Bibliography available on request.

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