

Specification

Differential and selective solid medium for the isolation of *Salmonella* and some *Shigella* species from clinical specimens, foods, etc.

Presentation

	Packaging Details	Shelf Life	Storage
20 Prepared Plates 90 mm with: 21 ± 2 ml	1 box with 2 packs of 10 plates/pack. Single cellophane..	3 months	2-14 °C

Composition

Composition (g/l):	
Meat extract.....	5.00000
Peptone.....	5.00000
Lactose.....	10.00000
Bile salts.....	5.60000
Sodium citrate.....	10.00000
Sodium thiosulfate.....	8.50000
Ferric citrate.....	1.00000
Brilliant green.....	0.00033
Neutral red.....	0.02500
Agar.....	15.00000

Description /Technique

Description:

SS Agar is a highly selective agar used for the isolation of *Salmonella* and *Shigella* species from very contaminated samples. Selectivity is obtained by a high concentration of bile salts and brilliant green, which inhibits the growth of Gram positive bacteria. The growth of other Gram negative flora is highly repressed due to the presence of citrate and thiosulfate. Some coliforms may still grow on this medium. Differentiation between pathogenic species and coliforms is achieved by the colour change of the pH indicator (neutral red). Lactose fermenters produce a pink or red coloured medium and colonies, while non-fermenting species form colourless colonies and turn the medium yellow. Should any species produce H₂S, it is easily detected by the black precipitate of ferrous sulfide, which turn the colonies black.

The peptone and the meat extract are capable of inducing the growth of most pathogenic species, nevertheless some *Shigella* are very fastidious and may grow poorly.

Technique:

If it is suspected that organisms might have been damaged and the viability of the microorganisms is poor i.e. (processed food, faeces from the patients under antibiotic treatment, etc.) it is advisable to proceed with a prior enrichment in Selenite-Cystine Broth Base or Tetrathionate Broth Base. After enrichment, inoculate SS Agar plates heavily with the specimen and proceed in the same way as with other specimens on a less selective medium, such as Brilliant Green Agar or MacConkey Agar.

Incubate the inoculated plates at 37°C for 18-24 hours. The presumptive colonies should then be sub-cultured on differential media to be identified biochemically or serologically.

Appearance of the colonies after 24 hours on SS Agar:

- *Shigella*: Colourless, transparent and flat.
- *Salmonella* (Non H₂S producers): Colourless, transparent and flat.
- *Salmonella* (H₂S producers): Black or black centred, flat, with transparent borders.
- *Proteus*: Similar appearance as *Salmonella* colonies, but smaller in size.
- *Escherichia coli*: If they grow, they are small, convex and pink or red coloured.
- Coliforms (in general): Large, opaque, smooth and white or pink in colour.

Each laboratory must evaluate the results according to their specifications.

Quality control

Physical/Chemical control

Color : Pink pH: 6.9 ± 0.2 at 25°C

Microbiological control

10³-10⁴ CFU (Productivity test qualitative)/ 10⁴-10⁶ CFU (Selectivity)

Microbiological control according to ISO 11133:2014/A1:2018.

Analytical methodology according to ISO 11133:2014/A1:2018; A2:2020.

Aerobiosis. Incubation at 37 ± 1 °C, reading after 21 ± 3 h

Microorganism

Escherichia coli ATCC® 25922, WDCM 00013

Salmonella enterica ATCC® 13076, WDCM 00030

Shigella flexneri ATCC® 12022, WDCM 00126

Enterococcus faecalis ATCC® 29212, WDCM 00087

Salmonella typhimurium ATCC® 14028, WDCM 00031

Growth

Inhibited

Good. Colonies SH2 positive

Good. Colourless colonies w/o SH2

Partial Inhibition

Good. Colonies SH2 positive

Sterility Control

Incubation 48 h at 30-35 °C and 48 h at 20-25 °C: NO GROWTH.

Check at 7 days after incubation in same conditions.

Bibliography

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- GRAY, L.D. (1995) *Escherichia, Salmonella, Shigella* and *Yersinia*. In Murray, Baron, Pfaller Tenover & Tenover (eds) Manual Clinical Microbiology. 6th ed. ASM Washington DC.
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- ISO 11133:2014/ Adm 1:2018. Microbiology of food, animal feed and water. Preparation, production, storage and performance testing of culture media.
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Storage

Storage conditions: 2-14°C

Alternatively the plates may also be stored at the range of 2 - 25°C, with a proper performance of the medium, but some precautions must be taken into account:

-In the range of 2 - 8 °C avoid direct contact with surfaces that can freeze product.

-In the range of 15 - 25 °C, dehydration control must be taking in account.