



SS Agar, Modified

M1032

Intended Use:

SS Agar (Salmonella Shigella Agar) Modified is used for the selective isolation and differentiation of *Salmonella* and *Shigella* species from pathological specimens, suspected foodstuffs etc.

Composition**

Ingredients	g / L
Peptone	5.000
HM Peptone B #	5.000
Lactose	10.000
Bile salts mixture	5.500
Sodium citrate	10.000
Sodium thiosulphate	8.500
Ferric citrate	1.000
Brilliant green	0.00033
Neutral red	0.025
Agar	12.000
Final pH (at 25°C)	7.2±0.2

**Formula adjusted, standardized to suit performance parameters.

Equivalent to Beef extract.

Directions

Suspend 57.02 grams in 1000 ml purified/distilled water. Heat to boiling with frequent agitation to dissolve the medium completely. **DO NOT AUTOCLAVE OR OVERHEAT.** Overheating may destroy the selectivity of the medium. Cool to about 45-50°C. Mix and pour into sterile Petri plates.

Principle And Interpretation

Salmonella and *Shigella* are gram-negative, facultatively anaerobic, non-sporulating rods in the family *Enterobacteriaceae*. They are widely distributed in animals, infecting mainly the stomach and the intestinal tissues. SS Agar is recommended as differential and selective medium for the isolation of *Salmonella* and *Shigella* species from pathological specimens (1) and suspected foodstuffs (2,3,4,5) and for microbial limit test (6). SS Agar is a moderately selective medium in which gram-positive bacteria are inhibited by bile salts, brilliant green and sodium citrate.

Peptone and HM Peptone B provide essential growth nutrients. Lactose is the fermentable carbohydrate. Brilliant green, bile salts and thiosulphate selectively inhibit gram-positive and coliform organisms. Sodium thiosulphate is reduced by certain species of enteric organisms to sulphite and H₂S gas. This reductive enzymatic process is attributed to thiosulphate reductase. Production of H₂S gas is detected as an insoluble black precipitate of ferrous sulphide, formed upon reaction of H₂S with ferric ions or ferric citrate, indicated by black centered colonies.

The high selectivity of Salmonella Shigella Agar allows the use of large inocula directly from faeces, rectal swabs or other materials suspected of containing pathogenic enteric bacilli. On fermentation of lactose by few lactose-fermenting normal intestinal flora, acid is produced which is indicated by change of colour from yellow to red by the pH indicator neutral red. Thus these organisms grow as red-pigmented colonies. Lactose non-fermenting organisms grow as translucent colourless colonies with or without black centers. *Salmonella* species exhibit colourless colonies with black centers resulting from H₂S production. *Shigella* species form colourless colonies, which do not produce H₂S. While using samples suspected of being exposed to treatments that might have damaged the viability of microorganisms due to processing of food materials or samples from patients under antibiotic treatment etc., previous enrichment in Selenite cystine Broth Base (M025) or Tetrathionate Broth Base (M032) is necessary. Inoculate SS Agar plates with the enriched culture. After incubation the suspicious colonies should be subcultured on differential media to be identified biochemically or serologically.

Type of specimen

Clinical: faeces, rectal specimen ; Suspected foodstuffs.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (7,8).

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (2,3,4,5).

Warning and Precautions

In Vitro diagnostic use. For professional use only. Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling clinical specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. The medium is highly selective and may be toxic to certain *Salmonella* or *Shigella* species. Hence it is recommended to use to inoculate plates of less inhibitory media parallel to SS Agar, such as Hektoen Enteric Agar (M467) or Deoxycholate Citrate Agar (M065) for easier isolation of *Shigella* species (9).

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Light yellow to pink homogeneous free flowing powder.

Gelling

Firm, comparable with 1.2% Agar gel.

Colour and Clarity of Prepared Medium

Reddish orange coloured clear to slightly opalescent gel forms in Petri plates.

Reaction

Reaction of 5.7% w/v aqueous solution at 25°C. pH : 7.2±0.2.

pH

7.00-7.40

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony
<i>Escherichia coli</i> ATCC 25922 (00013*)	50-100	fair	20-30%	pink with bile precipitate
# <i>Klebsiella aerogenes</i> ATCC 13048 (00175*)	50-100	fair	20-30%	cream pink
<i>Enterococcus faecalis</i> ATCC 29212 (00087*)	50-100	none-poor	≤10%	colourless
<i>Proteus mirabilis</i> ATCC 25933	50-100	fair-good	30-40%	colourless, may have black centre
<i>Salmonella</i> Choleraesuis ATCC 12011	50-100	good-luxuriant	≥50%	colourless with black centre
<i>Salmonella</i> Typhi ATCC 6539	50-100	good-luxuriant	≥50%	colourless with black centre
<i>Salmonella</i> Typhimurium ATCC 14028 (00031*)	50-100	good-luxuriant	≥50%	colourless with black centre
<i>Salmonella</i> Enteritidis ATCC 13076 (00030*)	50-100	good-luxuriant	≥50%	colourless with black centre
<i>Shigella flexneri</i> ATCC 12022 (00126*)	50-100	good	40-50%	colourless

* Corresponding WDCM numbers

(#) Formerly known as *Enterobacter aerogenes*

Storage and Shelf Life

Store between 10- 30°C in a tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (7,8).

Reference

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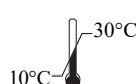
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