

## TECHNICAL DATA SHEET

# SALMONELLA-SHIGELLA (SS) AGAR

### ISOLATION OF *SALMONELLA* AND *SHIGELLA*

## 1 INTENDED USE

*Salmonella-Shigella* (SS) agar is used for the isolation of *Salmonellae* and *Shigellae* in fecal material. It can also be used as a second media of isolation in the context of standardized methods for the detection of *Salmonella*.

## 2 HISTORY

A number of authors, including Hormaeche, Surraco, Hardy, Rose, Mayfiels, Goeber, Pots and Caudill, successfully used S.S. agar for the isolation of *Salmonella* and *Shigella*. Schaub showed that the medium was useful for the production of hydrogen sulfide by the bacteria sought.

## 3 PRINCIPLES

SS Agar is a moderately selective medium in which Gram-positive bacteria are inhibited by bile salts, brilliant green and sodium citrate.

The high concentrations of citrate and sodium thiosulfate limit the development of coliform bacteria and prevent the invasion of the medium by *Proteus*.

Fermentation of lactose to acid is revealed by the formation of red colonies in the presence of neutral red. Lactose-negative bacteria form colorless colonies.

In the presence of thiosulfate and ferric citrate, hydrogen sulfide producers form colonies with a black center.

## 4 TYPICAL COMPOSITION

The composition can be adjusted in order to obtain optimal performance.

For 1 liter of media :

- Pancreatic digest of meat.....	5,0 g
- Meat extract.....	5,0 g
- Lactose.....	10,0 g
- Bile salts.....	8,5 g
- Sodium citrate.....	10,0 g
- Sodium thiosulfate.....	8,5 g
- Ferric ammonium citrate.....	1,0 g
- Neutral red.....	25,0 mg
- Brilliant green.....	0,33 mg
- Bacteriological agar.....	15,0 g

pH of the ready-to-use media at 25 °C : 7,0 ± 0,2.

## 5 PREPARATION

- Dissolve 63,0 g of dehydrated media (BK022) in 1 liter of distilled or demineralized water.
- Slowly bring to boiling, stirring with constant agitation until complete dissolution.
- Do not autoclave.
- Cool and maintain the media in a molten state at 44-47 °C.
- Pour into sterile Petri plates and let solidify on a cold, flat surface.

✓ **Reconstitution :**  
63,0 g/L

✓ **Sterilization :**  
Bring to a boil.

## 6 INSTRUCTIONS FOR USE

- Dry the plates in an incubator, covers partially removed.
- Inoculate by streaking the enrichment media used on the surface of the medium.
- Transfer in parallel the inoculum onto a another selective medium.
- Incubate at 37 °C for 24 to 48 hours.

✓ **Inoculation :**  
**Surface streaking**

✓ **Incubation :**  
**24 to 48 h at 37 °C**

## 7 RESULTS

*Salmonella* species not fermenting lactose form colorless, transparent colonies, with or without a black center (H<sub>2</sub>S production). *Shigella* are colorless.

Colonies of coliform bacteria are red or pink.

Suspected colonies are transferred to Kligler Iron Agar (BK034) or TSI Agar (BK059) for subsequent identification.

## 8 QUALITY CONTROL

**Dehydrated media :** pinkish powder, free-flowing and homogeneous.

**Prepared media :** orange-pink agar.

Typical culture response after 48 hours of incubation at 37 °C, qualitative method of inoculation :

Microorganisms		Growth	Characteristics
<i>Salmonella</i> Enteritidis	WDCM 00031	Good, score 2	Colorless colonies with a black center
<i>Salmonella</i> Typhimurium	WDCM 00030	Good, score 2	Colorless colonies with a black center
<i>Shigella flexneri</i>	WDCM 00125	Good, score 2	Colorless colonies
<i>Shigella sonnei</i>	WDCM 00127	Good, score 2	Red colonies
<i>Escherichia coli</i>	WDCM 00013	Partially inhibited, score 0-1	-
<i>Enterococcus faecalis</i>	WDCM 00087	Inhibited, score 0	-
<i>Staphylococcus aureus</i>	WDCM 00034	Inhibited, score 0	-

## 9 STORAGE / SHELF LIFE

**Dehydrated media :** 2-30 °C.

The expiration date is indicated on the label.

**Prepared media in plates (\*) :** 15 days at 2-8 °C, shielded from light.

(\*) Benchmark value determined under standard preparation conditions, following manufacturer's instructions.

## 10 PACKAGING

**Dehydrated media :**

500 g bottle ..... BK022HA

## 11 BIBLIOGRAPHY

Leifson, E.. 1935. New culture media based on sodium desoxycholate for the isolation of intestinal pathogens and for the enumeration of colon bacilli in milk and water. The Journal of Pathology and Bacteriology, **40** : 581-599.

Taylor, W.I., and Harris, B.. 1965. Isolation of shigellae. II. Comparison of plating media and enrichment broths. American Journal of Clinical Pathology, **44** : 476-479.

Isenberg, H.D., Kominos, S., and Siegel, M.. 1969. Isolation of Salmonellae and Shigellae from an Artificial Mixture of Fecal Bacteria. Applied Microbiology, **18(4)** : 656.-659.

Horwitz, W.. 1980. Official methods of analysis of the Association of Official Analytical Chemists, Washington DC.

NF EN ISO 6579-1. Avril 2017. Microbiologie de la chaîne alimentaire - Méthode horizontale pour la recherche, le dénombrement et le sérotypage des Salmonella - Partie 1 : recherche des Salmonella spp..

## 12 ADDITIONAL INFORMATION

---

The information provided on the labels take precedence over the formulations or instructions described in this document and are susceptible to modification at any time, without warning.

Document code : SALMONELLA SHIGELLA AGAR\_ENv8  
Creation date : 03-2003  
Updated : 02-2018  
Origin of revision : Bibliography.