Instaprep® Salmonella Shigella Agar

Intended Use
Instaprep® Salmonella Shigella Agar is used for differential and selective isolation of *Salmonella* and some *Shigella* species from pathological specimens and suspected food stuffs.

Summary
Salmonella Shigella (SS) Agar is the standard medium for the isolation of *Salmonella* and *Shigella* spp. from pathological specimens such as stools and suspected foodstuff and for microbial limit test. It is a highly selective and differential medium. Cultivation and isolation of bacteria from pathological samples is many a times key to the identification of the underlying infections. Availability of microbiology testing and such procedures being available in routine laboratories has been limited due to the availability of dehydrated media which can be put to use only after substantial procedural and preparatory requirements. Instaprep® Salmonella Shigella Agar media are ready to use / ready to pour and fill this long felt need using a unique proprietary technology for routine microbiological testing.

Principle
Instaprep® Salmonella Shigella Agar ready to pour media are pre-sterilised media with standard proven formulations. The pouched media only need to be kept in boiling (100°C) water for ten minutes and they become ready to pour into sterile plates. A result of Tulip's long research the Instaprep® Salmonella Shigella Agar pouched media accord flexibility to the laboratories, thereby avoiding laborious preparatory steps and wastage. Instaprep® Salmonella Shigella Agar media also help laboratories to set up cultures on a random basis and not to be restricted to batching of cultures. As compared to pre-poured plates and dehydrated media, variability, contamination and wastage is also avoided.
Agar contains lactose to differentiate between the lactose fermenting coliforms from the lactose non-fermenting *Salmonella* and *Shigella* species. The bile salt in combination with brilliant green inhibit Gram-positive bacteria, most coliform bacteria and prevent swarming phenomenon of *Proteus* spp.Sodium thiosulphate is reduced by certain species of enteric bacteria to sulphite and H_2S gas. H_2S reacts with ferric citrate and forms a black insoluble precipitate of ferrous sulphide in the centers of the colonies. Neutral red and brilliant green are present as pH indicators. The high selectivity of Salmonella Shigella (SS) Agar allows the use of large inocula directly from faeces, rectal swabs and other materials suspected of containing pathogenic enteric bacilli. The poured medium is a clear to slightly opalescent, very light buff to pink colour with a pH at 7.0 ± 0.2.

Reagent
Instaprep® Salmonella Shigella Agar is a ready to pour sterilized pouched media for microbiological applications such as cultivation / isolation / selective growth /susceptibility tests.

Formula
Ingredients	**g/L**
Lactose	10.0
Sodium Citrate	10.0
Bile Salts Mixture	8.5
Sodium Thiosulphate	8.5
Peptic Digest of Animal Tissue	5.0
Beef Extract	5.0
Ferric Citrate	1.0
Neutral Red	0.025
Brilliant Green	0.00033
Agar	15.0
Final pH (at 25°C ± 2°C)	7.0 ± 0.2

*Adjusted to suit performance parameters.

Additional Material Required
Water bath (250 mL beaker) at 100°C, vertical laminar air flow/ biosafety hood with Bunsen burner, forceps/ tongs, sterile petriplates (disposable/ glass), scissors, disinfectant (70% alcohol), absorbent sterile gauze, plastic/ glass/ wire rod for hanging pouches in water bath.

Procedure

1. Retrieve the required number of pouches from the carton.
2. Gently squeeze the gelled media to the bottom of the (Dip side) pouch, upto 'SQZ' mark.
3. Hang the pouches vertically using a hanging rod in a boiling water bath (at 100°C) with the 'DIP' side into the water and the water level upto the 'MAX' mark, for 10 minutes. Ensure that the heat source is not directly applied to the pouch. Retrieve the pouches after 10 minutes. In case rod hanger is not used for the pouches, remove the pouches using forceps/tongs. After retrieving the pouch, it should be dried diligently with gauze and then disinfected. Any residual water from the water bath should not be allowed to drip on to the poured plate to avoid contamination.
4. Wipe dry the pouch corner at the 'CUT' mark and disinfect with 70% isopropyl alcohol (IPA).
5. Cut the pouch across the 'CUT' mark with disinfected scissors.
6. For 15 mL pouches, squeeze and pour out media aseptically into a sterile 90 mm (diameter) petriplate.
7. While pouring the media take care not to splash or form air bubbles.
8. Cover the petriplate and allow the poured media to set.
9. The poured plate is now ready to use.
10. The samples should be collected and processed aseptically before plating.

Quality Control

Appearance: 15 mL pouch with intact seal and white crystalline powder of selective inhibitor.
Appearance of the poured plate: Slight opalescent, light pink coloured firm gel forms in petriplates.
Cultural Response: Cultural characteristics observed after an incubation of 18-24 hours at 30°C-35°C.

Organisms (ATCC)	**Growth**	**Colour of Colony**
Salmonella enterica subsp. *enterica* serovar *Typhimurium* (14028)	Good	Colourless with black centres
Shigella flexneri serotype 2b (12022)	Good	Colourless
Enterococcus faecalis (29212)	Partial Inhibition	Colourless
Escherichia coli (25922)	Complete Inhibition	-

Note: For good growth- Growth obtained on the test media should not differ by a factor greater than 2 from the calculated value for a standardized inoculum. For inhibition no growth of the test microorganism should occur. Inoculum cfu for good growth is 10-100. Inoculum cfu for Inhibition is >100.

Remarks

1. The temperature of water bath must be at 100°C to liquify the media. Cooler water baths will provide lumpy, uneven media.
2. Since all agar-based media solidify rapidly, it is important that minimum time be lost between retrieval of the pouch from boiling water bath and pouring aseptically into the sterile plates. This will produce evenly surfaced medium.
3. Good laboratory practices and hazard precautions must be observed at all times.
4. 15 mL media is sufficient for the standard 90 mm petriplates. In case smaller petriplates are being used more number of plates can be poured with a single pouch, proportionately.

Storage and Stability

1. Store the pouches at room temperature (15°C-25°C).
2. Stability of the unopened pouch is as per the expiry date mentioned on the label.

Warranty

This product is designed to perform as described on the label and package insert. The manufacturer disclaims any implied warranty of use and sale for any other purpose.

Reference

1. Basic Laboratory Procedures in Clinical Bacteriology, J. Vandepitte, K. Engbaek, P. Piot, C.C. Heuck, W.H.O. Geneva, 1991.
2. Diagnostic Microbiology, Bailey & Scott, 9th Ed., Mosby 1994, Ellen Jo Baron, Lance R. Peterson.
3. Practical Medical Microbiology, Mackie & McCartney, Vol. 1, Microbial Infections, 13th Ed., Churchill Livingston 1978, Edited J.P. Duguid, B.P. Marmion, R.H.A. Swain.

4. Practical Medical Microbiology, Mackie & McCartney, Vol. 2, 13th Ed., Churchill Livingston 1989, Edited by J.G. Collee Duguid, A.G. Fraser, B.P. Marmion.
5. Handbook of Microbiological Media, Ronald M. Atlas, Lawrence C. Parks, 2nd Ed., 1997.
6. Data on file: Microxpress®, A Division of Tulip Diagnostics (P) Ltd.

Product Presentation:

Cat. No.	Product Description	Pack Size
203090090015	Instaprep® Salmonella Shigella Agar	20 x 15 mL