



PRODUCT SPECIFICATION SHEET

S. S. Agar Modified / Salmonella Shigella Agar (Modified) DM237

Intended Use

Salmonella Shigella Agar (Modified) is used for the selective isolation of Salmonellae from faeces, urine, sewage and other materials.

Product Summary and Explanation

The culture media that have been developed for the selection and differentiation of enteric microorganisms from clinical and nonclinical materials inhibit the growth of gram-positive species to a varying degree due to the presence of either pure bile salts, mixtures of bile salts or dyes. SS Agar and Salmonella Shigella Agar (modified) are examples of media used in the plating of samples for the detection of enteric pathogens that contain bile salt mixtures. This formulation is essentially a modification of the Desoxycholate Citrate Agar described by Leifson.⁽¹⁾ Salmonellosis continues to be an important public health problem worldwide. Infection with non-typhi *Salmonella* often causes a mild, self-limiting illness. Typhoid fever, caused by *Salmonella typhi*, is characterized by fever, headache, diarrhoea, abdominal pain, and can result in fatal respiratory, hepatic, and or neurological damage.⁽²⁾ This infection can result from the consumption of raw, undercooked, or improperly processed foods contaminated with *Salmonella* spp. Shigellosis, caused by *Shigella* spp., is an intestinal illness characterized by abdominal pain, fever, and watery diarrhea. When associated with outbreaks, shigellosis is usually transmitted through contaminated food and/or water. Salmonella Shigella Agar is a modification of the Desoxycholate Citrate Agar described by Leifson.⁽³⁾ Salmonella Shigella Agar is superior to a number of other media for the isolation of *Salmonella* spp. and *Shigella* spp.⁽⁴⁾ Salmonella Shigella Agar is recommended for testing clinical specimens and food testing for the presence of *Salmonella* spp. and some *Shigella* spp.^(2,5,6)

Principles of the Procedure

In this media, Beef Extract and Peptic digest of animal tissue provide sources of nitrogen, carbon, and vitamins required for organism growth. Lactose is the carbohydrate present in Salmonella Shigella Agar. Bile Salts, Sodium Citrate and Brilliant Green inhibit gram-positive bacteria, most coliform bacteria, and inhibit swarming *Proteus* spp., while allowing *Salmonella* spp. to grow. Sodium Thiosulfate and Ferric Citrate permit detection of hydrogen sulfide by the production of colonies with black centers. Neutral Red is the pH indicator.

Formula / Liter

Ingredients	Gms / Liter
Beef extract	5.00
Peptic digest of animal tissue	5.00
Lactose	10.00
Bile salts mixture	5.50
Sodium citrate	10.00
Sodium thiosulphate	8.50
Ferric citrate	1.00
Brilliant green	0.00033
Neutral red	0.025
Agar	12.00
Final pH: 7.2 ± 0.2 at 25°C	
Formula may be adjusted and/or supplemented as required to meet performance specifications	





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Precautions

1. For Laboratory Use only.
2. IRRITANT. Irritating to eyes, respiratory system, and skin.

Directions

1. Suspend 57.02 grams of the medium in one liter of distilled water.
2. Heat if necessary, to dissolve the medium completely.
3. Autoclave at 121°C , 15 psi pressure, for 15 minutes / validated cycle.
4. Mix well and pour into sterile Petri plates.

Quality Control Specifications

Dehydrated Appearance	Light yellow to pink homogeneous free flowing powder
Prepared Medium	Reddish orange coloured clear to slightly opalescent gel forms in Petri plates
Reaction of 5.7% Solution	pH : 7.2 ± 0.2 at 25°C
Gel Strength	Firm, comparable with 1.2% Agar gel

Expected Cultural Response: Cultural characteristics observed after an incubation at 35 - 37°C for 18 - 48 hours.

Sr. No.	Organisms	Results to be achieved			
		Inoculum (CFU)	Growth	Recovery	Colour of colony
1.	<i>Escherichia coli</i> ATCC 25922	50 -100	Fair	20-30%	pink with bile precipitate
2.	<i>Enterobacter aerogenes</i> ATCC 13048	50 -100	Fair	20-30%	cream pink
3.	<i>Enterococcus faecalis</i> ATCC29212	50-100	None-poor	<=10%	colourless
4.	<i>Proteus mirabilis</i> ATCC 25933	50-100	Fair-good	30-40%	colourless, may have black centre
5.	<i>Salmonella choleraesuis</i> ATCC 12011	50-100	Good-luxuriant	>=50%	colourless with black centre
6.	<i>Salmonella typhi</i> ATCC 6539	50-100	Good-luxuriant	>=50%	colourless with black centre
7.	<i>Salmonella typhimurium</i> ATCC 14028	50-100	Good-luxuriant	>=50%	colourless with black centre
8.	<i>Salmonella enteritidis</i> ATCC 13076	50-100	Good-luxuriant	>=50%	colourless with black centre
9.	<i>Shigella flexneri</i> ATCC 12022	50-100	Good	40-50%	colourless

The organisms listed are the minimum that should be used for quality control testing.

Test Procedure

1. Use standard procedures to obtain isolated colonies from specimens.
2. A non-selective medium should also be streaked to increase the chance of recovery when the population of gram-negative organisms is low and to provide an indication of other organisms present in the specimen.





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3. Incubate plates, protected from light, at $35 \pm 2^{\circ}\text{C}$ for 18-24 hours.
4. If negative after 24 hours, reincubate an additional 24 hours.
5. For isolation of *Salmonella* spp. and *Shigella* spp. from clinical specimens, inoculate fecal samples and rectal swabs onto one quadrant of Salmonella Shigella Agar, streak for isolation.
6. Incubate plates at 35°C , and examine after 24 and 48 hours for colonies resembling *Salmonella* spp. or *Shigella* spp.

Results

1. Enteric organisms are differentiated by their ability to ferment lactose. *Salmonella* spp. and *Shigella* spp. are non-lactose fermenters and form colorless colonies on Salmonella Shigella Agar.
2. H_2S positive *Salmonella* spp. produce black-center colonies.
3. Some *Shigella* spp. are inhibited on Salmonella Shigella Agar.
4. *E. coli* produces pink to red colonies and may have some bile precipitation.

Storage

Store the sealed bottle containing the dehydrated medium at $10 - 30^{\circ}\text{C}$. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light.

Expiration

Refer to the expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if the appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitations of the Procedure

1. Salmonella Shigella Agar is highly selective and not recommended as the primary isolation of *Shigella*.^(1,2,6) Some *Shigella* spp. may be inhibited.
2. A few non-pathogenic organisms may grow on Salmonella Shigella Agar. These organisms can be differentiated by their ability to ferment lactose and other confirmatory tests.

Packaging

Product Name : Salmonella Shigella Agar (Modified)

Product Code : DM237

Available Pack sizes : 100gm / 500gm

References

1. Leifson. 1935. J. Pathol. Bacteriol. 40:581.
2. P. R. Murray, E. J. Baron, M. A. Pfaller, F. C. Tenover, and R. H. Tenover (eds.). Manual of clinical microbiology, 6th ed. American Society for Microbiology, Washington, D. C.s
3. 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. J. Pathol. Bacteriol 40:581.
4. Rose, H. M., and M. H. Kolodny. 1942. The use of SS (*Shigella-Salmonella*) Agar for the isolation of Flexner Dysentery bacilli from the feces. J. Lab. Clin. Med. 27:1081-1083.
5. Isenberg, H. D. (ed.). 1992. Interpretation of aerobic bacterial growth on primary culture media, Clinical microbiology procedures handbook, vol. 1 p. 1.61-1.67. American Society for Microbiology, Washington, D.C.
6. Vanderzant, C., and D. F. Splittstoesser (eds.). 1992. Compendium of methods for the microbiological examination of foods, 3rd ed. American Public Health Association, Washington, D.C.





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Further Information

For further information please contact your local MICROMASTER Representative.



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