



PRODUCT SPECIFICATION SHEET

Xylose-Lysine Deoxycholate Agar Plate (RP033H)

Intended Use

Xylose-Lysine Deoxycholate Agar (RP033H) is recommended for selective isolation and enumeration of *Salmonella typhi* and other *Salmonella* species from pharmaceutical products using microbial limit testing in compliance with harmonized methodology of USP/EP/BP/JP.

Product Summary and Explanation

The Enterobacteriaceae is a large family of Gram-negative bacteria that includes, along with many harmless symbionts, many of the more familiar pathogens, such as *Salmonella*, *Escherichia coli*, *Yersinia pestis*, *Klebsiella* and *Shigella*. Other disease-causing bacteria in this family include *Proteus*, *Enterobacter*, *Serratia*, and *Citrobacter*. They normally inhabit the intestines of humans and animals.⁽¹⁾ Human *Salmonella* infections are most commonly caused by ingestion of food, water or milk, contaminated by human or animal excreta.⁽²⁾ A wide variety of media have been developed to aid in the selective isolation and differentiation of enteric pathogens. Due to the large numbers of different microbial species and strains with varying nutritional requirements and chemical resistance patterns, investigators have developed various formulae to meet general as well as specific needs relative to isolation and identification of the microorganisms. Xylose Lysine Deoxycholate Agar is a selective as well as differential medium formulated by Taylor⁽³⁻⁷⁾ for the isolation and identification of enteric pathogens especially Shigellae from stool samples. This medium is also employed for pharmaceutical testing and non-sterile product testing for the detection (or absence) of *Salmonella* after enrichment in Rappaport Vassiliadis Salmonella Enrichment Broth (DM1405H) in accordance with the harmonized method of USP/EP/BP/JP/IP.⁽⁸⁻¹²⁾

Principles of the Procedure

Xylose-Lysine Deoxycholate Agar contains yeast extract which is a source of nitrogen, carbon, and vitamins required for organism growth. Deoxycholate, ferric ammonium citrate and sodium thiosulphate are selective agents that inhibit gram-positive microorganisms. Xylose, lactose, and sucrose are fermentable carbohydrate sources. Almost all the enteric bacteria except Shigellae, ferment xylose fermented which enables the differentiation of Shigellae from Salmonellae. Salmonellae metabolize the xylose and decarboxylate lysine and thus changing the pH to alkaline and mimic Shigellae reaction. Lactose and sucrose are added in excess to produce acid and hence non-pathogenic H₂S producers do not decarboxylate lysine thereby preventing Shigellae reaction by lysine positive coliforms. Sodium thiosulphate prevents the desiccation of these compounds during storage by reactivating sulphur containing compounds. It also forms the substrate for enzyme thiosulphate reductase, which breaks it to form H₂S. Thiosulphate and ferric ammonium citrate forms H₂S indicator system. Sodium thiosulphate is also inactivator of halogens, mercurial and aldehyde and can minimize its toxicity in the testing sample, if any during microbial limit tests. Sodium chloride maintains the osmotic equilibrium in this medium. Phenol red is the pH indicator.

Formula / Liter

Ingredients	Gms / Liter
Xylose	3.50
L-Lysine	5.00
Lactose monohydrate	7.50
Sucrose	7.50
Sodium chloride	5.00
Yeast extract	3.00
Phenol red	0.08
Sodium deoxycholate	2.50
Sodium thiosulphate	6.80
Ferric ammonium citrate	0.80
Agar	13.50
Final pH: 7.4 ± 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.

Product Deterioration

Do not use plates if they show evidence of microbial contamination, discoloration, drying, cracking or other signs of deterioration.

Quality Control Specifications

Appearance	Sterile Xylose-Lysine Deoxycholate Agar Plate Agar in 90mm plates
Colour	Red coloured clear to slightly opalescent gel
Reaction	pH : 7.4 ± 0.2 at 25°C
Quantity of medium	20-22ml of medium in 90mm plates

Sterility Check: Passes release criteria.

Expected Cultural Response:

Sr. No.	Organisms	Results to be achieved					
		Inoculum (CFU)	Growth	Observed Lot value (CFU)	Recovery	Colour of colony	Incubation Temperature
1.	Growth Promoting + Indicative						
2.	<i>Salmonella Typhimurium</i> ATCC 14028	50-100	good-luxuriant	25 -100	≥50 %	pink-red with bile precipitate	18 -24 hrs
3.	<i>Salmonella Abony</i> NCTC 6017	50-100	good-luxuriant	25 -100	≥50 %	pink to red	18 -24 hrs
4.	Additional Microbiological Testing						
5.	<i>Escherichia coli</i> NCTC 9002	50-100	fair	10 -30	20 -30 %	yellow	18 -72 hrs
6.	<i>Escherichia coli</i> ATCC 8739	50-100	fair	10 -30	20 -30 %	yellow	18 -72 hrs
7.	<i>Enterobacter aerogenes</i> ATCC 13048	50-100	fair	10 -30	20 -30 %	yellow	18 -72 hrs
8.	<i>Staphylococcus aureus</i> ATCC 25923	≥10 ³	inhibited	0	0 %	--	≥24 hrs
9.	<i>Staphylococcus aureus</i> ATCC 6538	≥10 ³	inhibited	0	0 %	--	≥24 hrs

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

Test Procedure

Refer to appropriate references for standard test procedures. This medium can be used in spread or pour plate procedures, with or without an overlay. In addition, this medium can be used as an over layer for spread plates to both prevent swarming colonies and to provide semi-anaerobic conditions that suppress the growth of non-fermentative gram-negative organisms. Stab inoculation procedures can also be used with this medium.



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Results

1. Degradation of fermentable sugars proceed concurrently and generates acids, which cause pH indicator to give various shades of colour, causing a colour change in the colonies and in the medium from red to yellow on prolonged incubation.
2. Hydrogen sulfide production results in colonies with black centers under alkaline conditions, which can be inhibited by acid production by carbohydrate fermentation. Alkaline condition causes the colour of the medium to change back to red.
3. This medium is an ideal medium for screening samples containing mixed flora of enteric pathogens as recovery of *Salmonellae* and *Shigellae* is not conspicuous by even profuse growth of other species.^(13,14)

Storage

On receipt, store plates at 20-25°C.

Expiration

Refer to the expiration date stamped on the pack. Prepared plates stored in their original sleeve wrapping at 20-25°C until just prior to use may be inoculated up to the expiration date and incubated for recommended incubation times.

Product Disposal

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

Limitations of the Procedure

1. When used in the pour plate procedure, the medium should be freshly prepared, tempered to 47°C, and used within 3 hours.
2. For identification, organisms must be in pure culture. Morphological, biochemical and/or serological tests should be performed for final identification.

Consult appropriate texts for detailed information and recommended procedures.

Packaging

Product Name : Xylose-Lysine Deoxycholate Agar Plate

Product Code : RPO33H

Available Pack sizes : Pack of 10 plates

References

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Further Information

For further information please contact your local MICROMASTER Representative.



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