



PRODUCT SPECIFICATION SHEET

XLT4 Agar Base (DM1007)

Intended Use

XLT4 Agar Base (DM1007) is recommended for selective isolation of *Salmonella* species other than *Salmonella typhi*.

Product Summary and Explanation

Salmonella is a genus of gram-negative enterobacteria commonly implicated in foodborne illness and is the causative agent of typhoid and paratyphoid fever. Although most *Salmonella* cannot be distinguished by biochemical characteristics, one serotype, namely *S. typhi* produce only a trace amount of hydrogen sulphide and is less active biochemically than the more common serotypes.⁽¹⁾

Numerous media have been developed for isolating and differentiating enteric pathogens. The majority were designed to recover a broad spectrum of enteric pathogens.⁽¹⁾ Consequently, overgrowth of nuisance or contaminating organisms can be a major problem when recovery of a specific organism or species is desired. This is particularly true for *Salmonella* isolation media where overgrowth of *Proteus*, *Providencia* and *Pseudomonas* can vividly interfere with the detection and isolation of *Salmonella*.⁽²⁾ In 1990, XLT4 Agar Base was formulated by Miller^(3,4) and Tate,⁽⁵⁾ for isolating *Salmonella* from faecally contaminated farm samples, which contains other bacteria as well. The authors established the selectivity of XLT4 Agar using pure cultures of a variety of enteric organisms. In follow-up studies, Miller and Tate reported that XLT4 Agar significantly improved the recovery of non-typhi *Salmonella* from chicken and farm environmental drag-swab samples. XLT4 Agar Base enhances the recovery of *Salmonella* species other than *Salmonella typhi*.^(3, 6-9)

Principles of the Procedure

XLT4 Agar Base contains proteose peptone is a source of carbon, nitrogen and other essential amino acids and growth factors. Yeast extract supplies nitrogenous requirements, vitamins and co factors required for growth. The sugars namely lactose, saccharose and xylose are the fermentable carbohydrates. Differentiation of *Salmonella* from other organisms that also grow on this medium is based on fermentation of xylose, lactose and sucrose, decarboxylation of lysine and the production of hydrogen sulfide. H₂S indicator system, consisting of sodium thiosulphate and ferric ammonium citrate is included for the visualization of the hydrogen sulphide produced, resulting in the formation of colonies with black centers. Sodium thiosulfate is added as a source of inorganic sulfur. The non-pathogenic H₂S producers do not decarboxylate lysine; therefore, the acid reaction generated by them prevents the blackening of the colonies.⁽¹⁰⁾ Sodium chloride maintains the osmotic balance of the medium. Phenol red is added as an indicator of pH changes resulting from fermentation and decarboxylation reactions. XLT4 Agar is both selective and differential. Tergitol-4 (MS102) inhibits growth of non-*Salmonella* organisms. Presumptive *Salmonella* colonies should be confirmed by performing biochemical tests.

Formula / Liter

Ingredients	Gms / Liter
Proteose peptone	1.60
Yeast extract	3.00
L-Lysine	5.00
Xylose	3.75
Lactose	7.50
Saccharose	7.50
Ferric ammonium citrate	0.80
Sodium thiosulphate	6.80
Sodium chloride	5.00
Phenol red	0.08
Agar	18.00
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.

Directions

1. Suspend 59.03 grams of the medium in one liter of distilled water.
2. Add 4.6 ml XLT4 Supplement (MS102).
3. Heat to boiling to dissolve the medium completely.
4. DO NOT AUTOCLAVE OR OVERHEAT.
5. Mix well and pour in sterile Petri plates.

Quality Control Specifications

Dehydrated Appearance	Light yellow to pink homogeneous free flowing powder
Prepared Medium	Red coloured clear to slightly opalescent gel forms in Petri plates
Reaction of 5.9% solution	pH 7.4 + 0.2 at 25°C
Gel Strength	Firm, comparable with 1.8% Agar gel

Expected Cultural Response: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours with added XLT4 Supplement (MS102).

Sr. No.	Organisms	Results to be achieved			
		Inoculum (CFU)	Growth	Recovery	Colour of Colony
1.	<i>Enterococcus faecalis</i> ATCC 29212	$\geq 10^3$	inhibited	0%	
2.	<i>Escherichia coli</i> ATCC 25922	50-100	fair-good	30-40%	yellow
3.	<i>Salmonella Enteritidis</i> ATCC 13076	50-100	good-luxuriant	$\geq 50\%$	red with black centers
4.	<i>Salmonella Typhimurium</i> ATCC 14028	50-100	good-luxuriant	$\geq 50\%$	red with black centers
5.	<i>Staphylococcus aureus</i> ATCC 25923	$\geq 10^3$	inhibited	0%	
6.	<i>Proteus mirabilis</i> ATCC 25933	50-100	none-poor	$\leq 10\%$	

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

Test Procedure

1. Inoculate a suitable *Salmonella* enrichment broth (such as Tetrithionate Broth) and incubate at 35°C for 18-24 hours.
2. Following enrichment, subculture onto XLT4 Agar. Streak for isolation.
3. Incubate plates aerobically at 35 ± 2°C. Examine for growth after 18-24 hours and 48 hours incubation.
4. Refer to appropriate references for standard test procedures.

Results

1. Typical *Salmonella* colonies (H₂S-positive) appear black or black-centered with a yellow periphery after 18-24 hours of incubation. Upon continued incubation, the colonies become entirely black or pink to red with black centers.
2. Colonies of H₂S-negative *Salmonella* strains appear pinkish-yellow.
3. Most *Citrobacter* colonies that grow on this medium are yellow without evidence of blackening.
4. Growth of *Enterobacter aerogenes* and *Escherichia coli* is markedly inhibited; colonies that do grow appear yellow without evidence of blackening.
5. Growth of *Proteus*, *Pseudomonas*, *Providencia*, *Alteromonas putrefaciens*, *Yersinia enterocolitica* and *Acinetobacter calcoaceticus* is markedly to completely inhibited on XLT4 Agar.
6. *Shigella* species are partially inhibited and colonies appear red.
7. Refer to appropriate references and test procedures for interpretation of results.



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Storage

Store the sealed bottle containing the dehydrated medium at 10 - 30°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. XLT4 Agar is intended for detecting and isolating *Salmonella* based on selectivity and colonial characteristics. Presumed *Salmonella* colonies must be confirmed by biochemical and/or immunological methods. Consult appropriate references for further information. 5-7
2. Non-*Salmonella* strains that are not completely inhibited on this medium may be encountered and must be differentiated from *Salmonella*. Consult appropriate references.
3. Freshly inoculated plates and plates held over several days may develop multicolored, metallic looking crystals/flecks on the surface. These crystals/flecks do not interfere with the performance of the medium.
4. Consult appropriate texts for detailed information and recommended procedures.

Packaging

Product Name : XLT4 Agar Base

Product Code : DM1007

Available Pack sizes : 500gm

References

1. Miller and Tate. 1990. The Maryland Poultryman April:2.
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3. Miller, Tate, Mallinson and Schemer. 1991. Poultry Science 70:2429.
4. Miller, Tate, Mallinson and Schemer. 1992. Poultry Science 71:398.
5. Tate, Miller and Mallinson. 1992. J. Food Prot. 55:964.
6. Miller R. G., Tate C. R., and Mallinson E. T. and Schemer J. A., 1991, Poultry science 70:2429
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8. Dusch H. and Altwegg M., 1994, Abstr. Annu. Meet. Am. Soc. Microbiol. C5:557
9. Dusch H. and Altwegg M., 1995, J. Clin. Microbiol. 33: 802 10. Taylor W. J., 1965, Am. J. Clin. Pathol., 44:471-475.

Further Information

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