XYLOSE LYSINE DEOXYCHOLATE (XLD) AGAR

PRODUCT:

Plated Media:

Xylose Lysine Deoxycholate Agar P2700

PURPOSE:

Xylose Lysine Deoxycholate (XLD) Agar is a moderately selective, differential media for the isolation of gram-negative enteric pathogens from fecal specimens. This media meets the U.S. Pharmacopeia (USP) standards in performing microbial examination of nonsterile products.

PRINCIPLE:

XLD was developed by Taylor[5] to improve the recovery of enteric pathogens, especially *Salmonella* and *Shigella* species, as compared to previously used selective media such as SS agar. Lactose, sucrose, and xylose are the fermentable carbohydrates present and phenol red is used as the pH indicator. Bacteria that ferment none of these sugars, e.g., *Shigella*, appear as red, translucent colonies. Yellow colonies indicate a rapid fermentation of lactose and acid pH, as demonstrated by *Escherichia coli*. Since *Salmonella* ferment xylose as readily as coliforms, a second differential mechanism, lysine decarboxylase, is utilized. Those organisms that ferment xylose as well as decarboxylate lysine exhaust the xylose rapidly and the lysine reaction causes a pH reversal to the alkaline reaction similar to *Shigella*. Lactose and sucrose are added in excess to prohibit this same reversion by lysine-positive coliforms. Sodium thiosulfate and ferric ammonium citrate are indicators of H_2S production only when alkaline conditions exist; *Salmonella* will, therefore, form red colonies with black centers in 24 hours while the H_2S production of *Citrobacter* and *Proteus* is greatly delayed. Sodium deoxycholate is added to inhibit gram-positive growth and to retard the growth of many strains of coliforms.

FORMULA: Approximate per liter formula.

Lactose	7.5 g
Sucrose	7.5
Xylose	3.5
L-Lysine	5.0
Sodium Thiosulfate	6.8
Sodium Chloride	5.0
Yeast Extract	3.0
Sodium Deoxycholate	2.5
Ferric Ammonium Citrate	0.8
Agar	13.5
Phenol Red	80.0 mg

Final pH 7.4 ± 0.2 at 25°C

PRECAUTIONS:*

For *in vitro* diagnostic use only. Observe approved biohazard precautions.

Storage: Upon receipt store at 2-8°C away from direct light. Media should not be used if there are signs of contamination, deterioration (shrinking, cracking, or discoloration), or if the expiration date has passed.

Limitations: Some *Salmonella* species, including *S. paratyphi A*, *S. choleraesuis*, *S. pullorum*, and *S. gallinarum*, may form red colonies without H_2S, thus resembling *Shigella* species.

Processing delays in excess of 2-3 hours (see “Specimen Collection”) of an unpreserved stool specimen greatly jeopardizes the recovery of most shigellas and many salmonellas. These organisms are very susceptible to the acidic change that occurs with a drop in temperature of the feces.[4]

Incubation times of over 48 hours may result in an alkaline reversion of normally acidic colonies, leading to false-positive results. However, optimum H_2S production is best visible between 24 and 48 hours. Misinterpretation may result unless the culture is held for 36-48 hours, but additional incubation may be misleading as well.

The few xylose-fermenting *Shigella* species and *Salmonella enteritidis*, which fail to decarboxylate lysine, may be missed on XLD agar, but would be detected on Hektoen Enteric Agar. Utilizing two media, each employing different differential systems, will optimize recovery of enteric pathogens by acting as a check for each other.

PROCEDURE:*

Specimen Collection: Information on specimen collection is found in standard reference material. In general, specimens should be protected from extremes of heat and cold and delivered to the laboratory within 2-3 hours. If there is a delay, suitable transport media such as Cary-Blair or Enteric Pathogen Transport must be used to maintain the viability of the organisms.

Method for Use: Prior to inoculation, the media should be brought to room temperature. Directly inoculate the stool or specimen from transport media onto the agar using standard microbiological procedures. Streak the inoculum so as to obtain isolated colonies. Incubate aerobically at 35°C for 18-48 hours (see "Limitations"). It is recommended that an enrichment broth such as GN or Selenite Broth be used and subcultured at the appropriate time to additional selective media to optimize recovery of *Salmonella* and *Shigella*.

Select red colonies with or without H_2S production for additional biochemical and serological testing.

Interpretation:

Salmonella paratyphimurium	Red to orange-pink colonies with black centers
Salmonella paratyphi	Red colonies with or without black centers
Salmonella species, *Edwardsiella tarda* *Proteus mirabilis*	Red colonies with black centers
Pseudomonas aeruginosa, Providencia species *Shigella* species	Red, transparent colonies
Escherichia coli, Klebsiella, Enterobacter, Citrobacter or *Serratia* species	Partial inhibition, Opaque yellow colonies

Materials Required but Not Provided: Standard microbiological supplies such as those commonly found in a microbiological laboratory are not provided.

QUALITY CONTROL:*

Microorganisms Used (ATCC#):	**Expected Results:**
Salmonella cholerasuis ssp. *cholerasuis* (14028)	Growth; Red colonies with black centers
Shigella flexneri (12022)	Growth; Red colonies
Escherichia coli (25922)	Inhibition, partial to complete
Enterococcus faecalis (29212)	Inhibition, partial to complete

User Quality Control: Check for signs of contamination and deterioration. XLD agar should appear translucent and red-orange in color.

BIBLIOGRAPHY:

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2. Murray, P.R., et al., *Manual of Clinical Microbiology*, 9th ed., American Society for Microbiology, Washington, D. C., 2007.
3. MacFaddin, J. F., *Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria*, vol. 1, Williams and Wilkins, Baltimore, 1985.
4. Sack, R. B., et al., *Cumitech 12*, American Society for Microbiology, Washington, D. C., 1980.
5. Taylor, W. I., and B. Harris, *Am. J. Clin. Pathol.*, 44: 476-479, 1965.
6. United States Pharmacopeia 30 - NF 25, Chapter 62, Microbial examination of nonsterile products: Tests for specified microorganisms, 2007.

*For more detailed information, consult appropriate references.

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