XLD AGAR (XYLOSE LYSINE DESOXYCHOLATE AGAR)

EUROPEAN PHARMACOPOEIA

CAT Nº: 1080

For the isolation of enteropathogenic bacteria, especially from the genus of *Shigella* and *Salmonella*

FORMULA IN g/l

Lactose Monohydrate	7.50	Yeast Extract	3.00
Sucrose	7.50	Sodium Desoxycholate	2.50
Sodium Thiosulfate	6.80	Ferric Ammonium Citrate	0.80
Sodium Chloride	5.00	Phenol Red	0.08
L-Lysine	5.00	Bacteriological Agar	13.50
Xylose	3.50		

Final pH 7.4 ± 0.2 at 25ºC

Salmonella typhimurium
ATCC 14028

PREPARATION

Suspend 55.2 grams of the medium in one liter of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. AVOID OVERHEATING. DO NOT AUTOCLAVE. Dispense into appropriate containers. The prepared medium should be stored at 8-15°C. The color is reddish-orange.

The dehydrated medium should be homogeneous, free-flowing and pink in color. If there are any physical changes, discard the medium.

USES

XLD AGAR was developed principally for isolating and differentiating Gram-negative enteric bacilli, particularly *Shigella* and *Salmonella*. It has been shown to be more effective than other enteric differential media.

The reactions that take place are the degradation of the three fermentable carbohydrates: xylose, lactose and sucrose, with the production of acid, manifested in the color change from red to yellow. Sodium thiosulfate serves as a reactive substance, with Ferric ammonium citrate as an indicator of the formation of hydrogen sulfide under alkaline conditions. Lysine allows the *Salmonella* group to be differentiated from the non-pathogens since, without it, salmonellae would quickly ferment the xylose and be indistinguishable from non-pathogenic species. Once the salmonellae consume the xylose, lysine is attacked via the enzyme, lysine decarboxylase, with a reversion to an alkaline pH which is similar to the *Shigella* reaction. The bacteria that decarboxylate the L-Lysine to cadaverine are indentified by the presence of a purple-red color around the colonies due to the elevation of the pH. Phenol red is the pH indicator. Yeast extract is the source of vitamins, particularly of the B-group essential for bacterial growth. Sodium chloride supplies essential electrolytes for transport and osmotic balance. Sodium desoxycholate is the selective agent inhibiting Gram-positive microorganisms. Bacteriological Agar is the solidying agent.

The European Pharmacopoeia recommends to inoculate and incubate *Escherichia coli* (indicative) as well as *Salmonella* at 30-35ºC during 18-48 hours.

It also recommends in Paragraph 2.6.13 "Microbiological examination of non-Sterile products: test for specified microorganisms" to subculture in this medium after incubation in Rappaport Vassiliadis Salmonella Enrichment Broth (Cat.1414), at 30-35ºC for 18-24 hours and incubate this medium at 30-35ºC for 18-48 hours.

Interpretation: The possible presence of *Salmonella* is indicated by the growth of well-developed red colonies, with or without black centers. This is confirmed by identification tests.

The product complies with the test if colonies of the types described are not present or if the confirmatory identification tests are negative.

CHARACTERISTICS OF COLONIES

ORGANISMS	COLONY CHARACTERISTISTICS
Arizona	Red and transparent with a black center
Citrobacter	Yellow and opaque. Can present a black center and clear edges
E.Coli, Enterobacter, Serratia	Yellow and opaque. Zone of yellow precipitation around the colonies
Edwardsiella	Red with a black center and clear edges
Klebsiella	Large, yellow, pale, mucoid and opaque. Zone of yellow precipitation around the colonies
Proteus mirabilis and *P.vulgaris*	Yellow, transparent, with clear edges. Black center especially *P.Mirabilis*
Proteus morganii and *P. rettgeri*	Red and transparent
Salmonella	Red, transparent with black centers and, if H2S is produced, yellow edges
Providencia and *Shigella*	Red and transparent

MICROBIOLOGICAL TEST

The following results were obtained in the performance of the medium from type cultures after incubation at a temperature of 30-35ºC and observed after 18-48 hours.

Microorganisms	Growth	Color Colony
Escherichia coli ATCC 25922	Partially inhibited	Yellow (precipitate)
Escherichia coli ATCC 8739	Partially inhibited	Yellow (precipitate)
**Salmonella typhimurium* ATCC 14028	Good	Clear Red (black center)
Shigella flexneri ATCC 12022	Good	Red
Staphylococcus aureus ATCC 25923	Inhibited	
Staphylococcus aureus ATCC 6538	Inhibited	

*According to European Pharmacopoeia Incubate at 30-35 ºC for 18-48 h

BIBLIOGRAPHY

Taylor, A. J. Clin. Path. 44:471. 1965. Taylor and Harris, A.J. Clin. Path. 44:476. 1965.

Rollender, W. U. Beckford; R.D. Belsky, B. Krostoff (1969) Comparison of Xylose Lysine desoxycholate agar and MacConkey agar for the isolation of Salmonella and Shigella from clinical specimens (tech. Bull. Reg. Med. Tech, 39 (1) 8-p)

European Pharmacopoeia. 7.0

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STORAGE

Once opened keep powdered medium closed to avoid hydration.