
XLD AGAR (ISO 6579-1)

DETECTION OF *SALMONELLA*

1 INTENDED USE

XLD (Xylose Lysine Desoxycholate) Agar is used mainly for the isolation of *Salmonella* and *Shigella* in food products. It is also used for the detection of *Salmonella* in animal health, (with mammals, birds and in animal production environments) and in water.

The typical composition corresponds to that defined in the standards NF EN ISO 6579-1, NF EN ISO 21527, NF EN ISO 19250, NF U47-100 to 102 and FD CEN/TR 15215-2 & 15215-3.

2 HISTORY

The medium was formulated by Taylor in order to increase the efficiency of recovery of pathogenic enterobacteria, particularly *Shigella* and other fastidious species which do not develop in other formulations containing highly toxic inhibitors.

3 PRINCIPLES

Sodium desoxycholate inhibits contaminating Gram-positive flora.

Xylose is fermented by practically all enteropathogenic bacteria, except for *Shigella* which are thus differentiated from the other species. After exhausting xylose, *Salmonella* decarboxylate lysine (via lysine decarboxylase) to cadaverine, causing the pH to rise. Colonies of salmonellae resemble those of shigellae in the medium having become basic.

The colonies formed are red in the presence of the indicator, phenol red.

The addition of lactose and sucrose to the medium enable coliform bacteria to decarboxylate lysine and thereby produce excess acidity, making the indicator turn yellow, favoring their differentiation.

In basic medium, pathogenic H₂S-producers reduce ferric ammonium citrate and cause a blackening due to the production of iron sulfide at the center of the colonies. Non-pathogenic bacteria which do not decarboxylate lysine acidify the medium, a result of sugar fermentation. The pH decrease prevents the colonies from blackening.

4 TYPICAL COMPOSITION

The composition can be adjusted in order to obtain optimal performance.

For 1 liter of media:

- Yeast extract	3,00 g
- L-Lysine hydrochloride	5,00 g
- Lactose	7,50 g
- Sucrose	7,50 g
- Xylose.....	3,75 g
- Sodium desoxycholate	1,00 g
- Sodium chloride	5,00 g
- Sodium thiosulfate.....	6,80 g
- Ferric ammonium citrate	0,80 g
- Phenol red	80 mg
- Bacteriological agar.....	12,50 g

pH of the ready-to-use media at 25 °C: 7,4 ± 0,2.

5 PREPARATION

- Dissolve 52,9 g of dehydrated media (BK168) in 1 liter of distilled or demineralized water.
- Slowly bring to boiling, stirring with constant agitation until complete dissolution.
- Cool rapidly and maintain in a molten state at 44-47 °C.
- Pour into sterile Petri plates and let solidify on a cold, flat surface.
- Dry the plates in an incubator, covers partially removed.

✓ **Reconstitution:**
52,9 g/L

✓ **Sterilization:**
Do not autoclave, pour rapidly into Petri plates.

Notes

Excessive heating or prolonged holding at 44-47°C may cause precipitation, so the colonies may furnish less clear-cut reactions.

The medium should present a clear aspect and orange-red.

6 INSTRUCTIONS FOR USE

- Re-inoculate a loop of enrichment media onto XLD plates prepared as above or using pre-poured plates (BM087).
- Incubate between 34 à 38°C for 24 ± 3 hours.

✓ **Inoculation:**
A loop of enrichment broth

✓ **Incubation:**
24 h between 34 to 38°C

Notes:

Incubation may be extended up to 48 hours in animal health.

A second media of choice, as in **COMPASS® Salmonella Agar** (BM066) should be inoculated in parallel.

In water microbiology, incubate at 36 ± 2 °C.

7 RESULTS

Salmonella present red colonies with or without a black center.

The appearance of the other colonies are as follows:

Characteristics	Microorganisms
Yellow colonies with a black center	<i>Escherichia coli</i> , <i>Citrobacter</i> , <i>Enterobacter</i> , <i>Proteus</i> , <i>Serratia</i> , <i>Klebsiella</i>
Red colonies, without black center	<i>Shigella</i> , <i>Providencia</i> , <i>Salmonella</i> Paratyphi A
Red colonies with a black center	<i>Salmonella</i> , <i>Edwardsiella</i>

See ANNEX 1: PHOTO SUPPORT.

8 QUALITY CONTROL

Dehydrated media: pinkish powder, free-flowing and homogeneous.

Prepared media: red-orange agar.

Typical culture response after 24 hours of incubation at 37 °C (NF EN ISO 11133):

Microorganisms	Growth	Characteristics
<i>Salmonella</i> Typhimurium WDCM 00031	Good, score 2	Red colonies with black center
<i>Salmonella</i> Enteritidis WDCM 00030	Good, score 2	Red colonies with black center
<i>Escherichia coli</i> WDCM 00013	Weak, score 0-1	Yellow colonies
<i>Enterococcus faecalis</i> WDCM 00087	Inhibited, score 0	-

9 STORAGE / SHELF LIFE

Dehydrated media: 2-30 °C.

Pre-poured media in Petri plates: 2-8 °C.

The expiration dates are indicated on the labels.

Prepared media in plates (*): 8 days at 2-8 °C.

(*) Benchmark value determined under standard preparation conditions, following manufacturer's instructions.

10 PACKAGING

Dehydrated media:

500 g bottle BK168HA

Pre-poured media in Petri plates (Ø 90 mm):

20 plates BM08708

11 BIBLIOGRAPHY

Taylor, W.I. 1965. Isolation of *Shigellae*. I. Xylose lysine agars; new media for isolation of enteric pathogens. American Journal of Clin. Path., **44(4)** : 471-475.

NF EN ISO 6579. Décembre 2002. Microbiologie des aliments. Méthode horizontale pour la recherche des *Salmonella* spp. Modifiée en Octobre 2007 par l'amendement A1 Annexe D : recherche de *Salmonella* spp. dans les matières fécales des animaux et dans des échantillons au stade de la production primaire.

NF EN ISO 21567. Mars 2005. Microbiologie des aliments. Méthode horizontale pour la recherche de *Shigella* spp.

FD/CEN/TR 15215-2. Avril 2006. Caractérisation des boues. Détection et dénombrement de *Salmonella* spp. dans les boues, les sols, les amendements du sol, les supports de culture et biodéchets. Partie 2 : Méthode par enrichissement en milieu liquide sélénite-cystine puis en milieu de Rapport-Vassiliadis pour la détermination semi-quantitative par la méthode du Nombre le Plus Probable (NPP).

FD/CEN/TR 15215-3. Avril 2006. Caractérisation des boues. Détection et dénombrement de *Salmonella* spp. dans les boues, les sols, les amendements du sol, les supports de culture et les biodéchets. Partie 3 : Présence/absence par enrichissement en milieu liquide peptone-novobiocine puis sur milieu Rapport-Vassiliadis.

NF U47-100. Juillet 2007. Méthodes d'analyse en santé animale. Recherche par l'isolement et identification de tout sérovar ou de sérovar(s) spécifié(s) de salmonelles dans l'environnement des productions animales.

NF U47-101. Novembre 2007. Méthodes d'analyse en santé animale. Isolement et identification de tout sérovar ou de sérovar(s) spécifié(s) de salmonelles chez les oiseaux.

NF U47-102. Janvier 2008. Méthodes d'analyse en santé animale. Isolement et identification de tout sérovar ou de sérovar(s) spécifié(s) de salmonelles chez les mammifères.

NF EN ISO 19250. Juin 2013. Qualité de l'eau. Recherche de *Salmonella* spp.

NF EN ISO 11133. Juillet 2014. Microbiologie des aliments, des aliments pour animaux et de l'eau. Préparation, production, stockage et essais de performance des milieux de culture.

ISO 6579-1. February 2017. Microbiology of the food chain - Horizontal method for the detection, enumeration and serotyping of *Salmonella* - Part 1: Detection of *Salmonella* spp.

NF EN ISO 6579-1/A1. March 2020. Microbiology of the food chain - Horizontal method for the detection, enumeration and serotyping of *Salmonella* - Part 1 : detection of *Salmonella* spp. - Amendment 1 Broader range of incubation temperatures, amendment to the status of Annex D, and correction of the composition of MSRV and SC

12 ADDITIONAL INFORMATION

The information provided on the labels take precedence over the formulations or instructions described in this document and are susceptible to modification at any time, without warning.

Document code : XLD ACC TO 6579_ENV11
Creation date : 11-2003
Updated : 06-2020
Origin of revision : Update according to the standard NF EN ISO 6579-1/A1

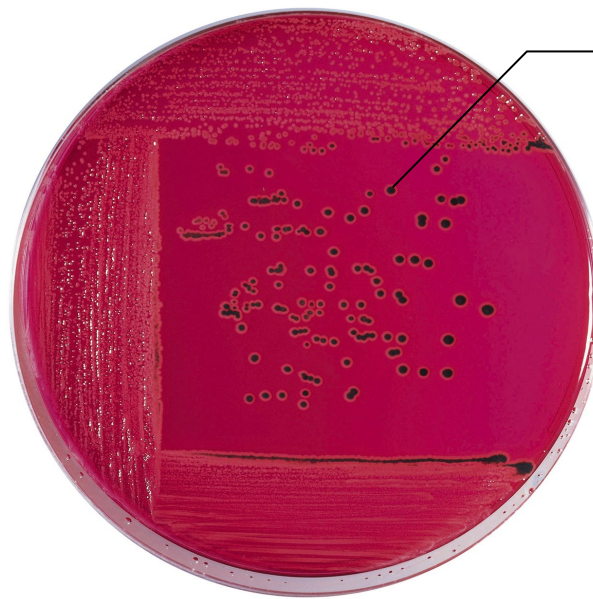
ANNEX 1: PHOTO SUPPORT

XLD Agar

Detection of *Salmonella*

Results:

Growth obtained after 24 hours of incubation at 37 °C.



***Salmonella* spp.**

Characteristic colonies:
red color with a black
precipitate in the center.