

Specification

Selective solid medium for the enumeration of enterobacteria, according to ISO standard 21528 and Pharmacopeial Harmonised Methods.

Presentation

10 Prepared bottles
Bottles 250 ml
with: 200 ± 5 ml

Packaging Details

1 box with 10 bottles 250 ml. metal non injectable cap.

Shelf Life

12 months

Storage

8-25°C

Composition

Composition (g/l):

Yeast extract.....	3.000
Peptone from gelatin.....	7.000
Salts bile.....	1.500
D(+) Glucose.....	10.000
Sodium chloride.....	5.000
Neutral red.....	0.030
Crystal violet.....	0.002
Agar.....	13.000

Description /Technique

Description

This medium is a modification of the Violet Red Bile Agar and the MacConkey Agar as described by Mossel et al. The addition of glucose to the Violet Red Bile Agar enhances both the growth of the most fastidious enterobacteria and the recovery of those having suffered from adverse conditions. Mossel himself realized that by removing the lactose and keeping the glucose, the medium's efficiency remained stable.

This medium can be used as a presumptive medium for E. coli (by fluorescent reaction) if before sterilization MUG is added.

Technique

Collect, dilute and prepare samples and volumes as required according to specifications, directives, official standard regulations and/or expected results.

Melt the medium contained in the bottles in a water bath (100°C) or in a microwave oven, avoiding overheating, before pouring into Petri dishes when cooled to room temperature.

Once solidified on a flat surface, Spread the plates by streaking methodology or by spiral method. Incubate the plates right side up aerobically at 35°C± 2,0 for 24 h.

(Incubation times longer than those mentioned above, different inoculation methods or different incubation temperatures may be required depending on the sample, on the specifications,...

This medium can be inoculated directly or after any enrichment broth)

after incubation, enumerate all the reddish-violet colonies that have appeared onto the surface of the agar, with a red-violet halo due to bile salts precipitation.

Presumptive isolation of E.coli or coliforms must be confirmed by further microbiological and biochemical tests.

calculate total microbial count per ml of sample by multiplying the average number of colonies per plate by the inverse dilution factor if streaked a diluted sample. Report results as Colony Forming Unit (CFU's) per ml or g along with incubation time and temperature, that enables to differentiate total coliforms and faecal coliforms.

Quality control**Physical/Chemical control**

Color : Violet-pink

pH: 7.4 ± 0.2 at 25°C

Microbiological control

Melting - pour plates - inoculation Practical range 100±20 CFU; Min. 50 CFU (Productivity) / 10⁴-10⁶ CFU (Selectivity)
Microbiological control according to ISO 11133:2014/ Adm 1:2018.

Aerobiosis. Incubation: 30-35°C. Reading at 24h (E.P.) / 37±1°C. Reading at 24 h (ISO)

Microorganism

Enterococcus faecalis ATCC® 19433, WDCM 00009
Salmonella typhimurium ATCC® 14028, WDCM 00031
Ps. aeruginosa ATCC® 9027, WDCM 00026
Escherichia coli ATCC® 8739, WDCM 00012
Staphylococcus aureus ATCC® 6538, WDCM 00032
Escherichia coli ATCC® 25922, WDCM 00013

Growth

Inhibited
Good (50%)- Red purple colonies - Biliar precipitate
Good
Good (50%)- Red purple colonies - Biliar precipitate
Inhibited
Good (50%)- Red purple colonies - Biliar precipitate
Note: results ATCC 8739/6538/9027 at 30-35 °C. Rest

Sterility Control

Incubation 48 hours at 30-35°C and 48 hours at 20-25°C: NO GROWTH
Check at 7 days after incubation in same conditions

Bibliography

- EUROPEAN PHARMA COEIA 8.0 (2014) 8th ed. § 2.6.13. Microbiological examination of non-sterile products: Test for specified microorganisms. Harmonised Method. EDQM. Council of Europe. Strasbourg.
- ISO Norma 21528-1: 2004. Microbiology of food and animal feeding stuffs - Horizontal methods for the detection and enumeration of Enterobacteriaceae - Part 1: Detection and enumeration by MPN technique with pre-enrichment.
- ISO 11133:2014/ Adm 1:2018. Microbiology of food, animal feed and water. Preparation, production, storage and performance testing of culture media.
- MOSSEL, D.A.A. (1985) Media for Enterobacteriaceae. Int. J. Food Microbiol. 2:27-35.
- MOSSEL, D.A.A., H. MENGERINK & H.H. SCHOLTS (1962) Use a Modified MacConkey Agar Medium for the selective growth and enumeration of all Enterobacteriaceae. J. Bact. 84:381.
- MOSSEL, D.A.A., M. VISER & A.M.R. CORNELISSEN (1963) The examination of foods for Enterobacteriaceae using a test of the type generally adopted for the detection of salmonellae. J. Appl. Bact. 26:444-452.
- MOSSEL, D.A.A. & M.A. RATTO (1970) Rapid detection of sub-lethally impaired cells of Enterobacteriaceae in dried foods. Appl. Microbiol. 20:273-275.
- PASCUAL ANDERSON, M^o R. (1992) Microbiología Alimentaria. Díaz de Santos, S.A. Madrid.
- USP 33 - NF 28 (2011) <62> Microbiological examination of non-sterile products: Test for specified microorganisms. Harmonised Method. USP Corp. Inc. Rockville. MD. USA.