

# MACCONKEY AGAR w/o CRYSTAL VIOLET

## INTENDED USE

Remel MacConkey Agar w/o Crystal Violet is a solid medium recommended for use in qualitative procedures for the taxonomic separation of the rapidly growing mycobacteria.

## SUMMARY AND EXPLANATION

MacConkey Agar w/o Crystal Violet is a modification of the original formulation described by MacConkey in 1905.<sup>1</sup> In 1974, investigators observed that certain strains of mycobacteria expected to grow on MacConkey Agar failed to do so.<sup>2</sup> Kubica et al. evaluated two formulations of MacConkey Agar for isolation of rapidly growing mycobacteria. MacConkey Agar w/o Crystal Violet provided for better recovery and differentiation of rapid growers, such as *Mycobacterium fortuitum-chelonae* complex.

## PRINCIPLE

This modification of MacConkey Agar has a higher concentration of bile salts and does not contain crystal violet. The elimination of crystal violet permits the growth of gram-positive cocci and acid-fast bacilli. Peptones provide nitrogenous compounds and amino acids necessary for bacterial growth. Sodium chloride maintains osmotic equilibrium. This test is valuable in the taxonomic separation of the rapidly growing acid-fast bacilli.<sup>3</sup> *M. fortuitum-chelonae* complex usually grows in 5-11 days and sometimes reveals a color change. Saprophytic strains of rapidly growing mycobacteria are inhibited by this medium.

## REAGENTS (CLASSICAL FORMULA)\*

|                      |        |                           |           |
|----------------------|--------|---------------------------|-----------|
| Gelatin Peptone..... | 17.0 g | Casein Peptone .....      | 1.5 g     |
| Lactose .....        | 10.0 g | Meat Peptone.....         | 1.5 g     |
| Bile Salts .....     | 5.0 g  | Neutral Red.....          | 0.04 g    |
| Sodium Chloride..... | 5.0 g  | Agar .....                | 12.0 g    |
|                      |        | Demineralized Water ..... | 1000.0 ml |

pH 7.4 ± 0.2 @ 25°C

\*Adjusted as required to meet performance standards.

## PROCEDURE

Follow established laboratory safety procedures when working with acid-fast cultures and specimens. Consult appropriate references when necessary for detailed procedural information on specimen processing, media inoculation, and identification of test isolates.<sup>3,4</sup>

1. Cultures to be tested should be grown for 7 days at 28°C in a suitable broth medium.
2. Inoculate plates with a 3 mm loopful of the broth culture and streak for isolation.
3. Incubate in ambient air at 28-35°C for 5 to 11 days. Do not incubate in CO<sub>2</sub>.
4. Examine plates for typical colony morphology and growth characteristics.

**Pour Tube:** Melt the pour tube in a boiling water bath and cool to 45-50°C. Mix and dispense into a sterile petri dish and proceed with the instructions above.

## QUALITY CONTROL

All lot numbers of MacConkey Agar w/o Crystal Violet have been tested using the following quality control organisms and have been found to be acceptable. Testing of control organisms should be performed in accordance with established laboratory quality control procedures. If aberrant quality control results are noted, patient results should not be reported.

### CONTROL

*Mycobacterium fortuitum* ATCC® 6841  
*Mycobacterium phlei* ATCC® 11758

### INCUBATION

Ambient, 5 days @ 33-37°C  
Ambient, 5 days @ 33-37°C

### RESULTS

Growth  
No growth

## LIMITATIONS

1. Strains of *Mycobacterium* other than *M. fortuitum-chelonae* complex may grow on MacConkey Agar w/o Crystal Violet, in particular, *Mycobacterium smegmatis*.<sup>3</sup>
2. *M. fortuitum-chelonae* complex may not reveal a color change when grown on this medium.<sup>3</sup>

## BIBLIOGRAPHY

1. MacConkey, A.T. 1905. J. Hyg. 5:333.
2. Kubica, G.P. and J. Vitvitsky. 1974. Appl Microbiol. 27:917-919.
3. Kent, P.T. and G.P. Kubica. 1985. Public Health Mycobacteriology-A Guide for the Level III Laboratory. U.S. Dept. of H.H.S. and CDC, Atlanta, GA.
4. Garcia, L.S. 2010. Clinical Microbiology Procedures Handbook. 3<sup>rd</sup> ed. ASM Press, Washington, D.C.

Refer to the front of Remel *Technical Manual of Microbiological Media* for **General Information** regarding precautions, product storage and deterioration, specimen collection, storage and transportation, materials required, quality control, and limitations.

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