

Protocol

TD-P Revision 3.0

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MUG Agar Protocol

Introduction

Escherichia coli (*E. coli*) is a bacterium found in the gastrointestinal tract of many organisms, including human beings. Most *E. coli* strains are harmless. However, some strains are known to cause deadly infections. Most *E. coli* also produce β-glucuronidase, which hydrolyses the substrate 4-methylumbelliferyl-β-D-glucuronide (MUG), resulting in the generation of 4-methylumbelliferone, a fluorescent product easily detectable under a UV light source. Thus, MUG agar is often used for the identification and isolation of *E. coli* strains found in food and pharmaceutical products. Here we describe how to prepare and use MUG agar plates.

Materials

Reagents and quantities needed for the medium:

- 20.0 g/L Casein peptone
- 2.0 g/L Meat extract
- 1.0 g/L Yeast extract
- 10.0 g/L Sorbitol
- 0.5 g/L Ammonium ferric citrate
- 0.1 g/L MUG (GoldBio Catalog # [MUG](#))
- 5.0 g/L Sodium chloride
- 2.0 g/L Sodium thiosulfate
- 0.025 g/L Bromothymol blue (GoldBio Catalog # [B-750](#))
- 1.12 g/L Deoxycholic acid sodium salt (GoldBio Catalog # [D-070](#))
- 13 g/L Agar
- Total quantity should be ~55 g/L.

Note: Store prepared medium below 8°C, at pH of 7.4 and protect from light. Store the dehydrated MUG powder at -20°C.

Note: The addition of sorbitol aids in identifying *E. coli* strains that can degrade sorbitol. The *E. coli* that can degrade sorbitol will result in yellow colonies. The strains that cannot degrade sorbitol, will appear as green colonies.

Note: Ammonium ferric citrate and sodium thiosulphate are added to help differentiate cultures that can produce hydrogen sulfide. These will appear as brown colonies.

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Method

1. Dissolve 55 g of mixture above in 1 L of molecular biology-grade water.
2. Autoclave at 121°C for 15 minutes.
3. Cool to 50°C.
4. Mix and pour into petri plates.
5. Inoculate the medium by spreading on the petri plates and incubate at ~37°C.
6. To check for *E. coli*, after a 24 hour incubation, check the plates under UV light at 360 nm.
7. The observation of a light blue fluorescence indicates the presence of *E. coli*. If fluorescence does not occur within 24 hours, continue to incubate for another 24 hours and check for blue fluorescence again.

Associated Products

- [MUG \(GoldBio Catalog # MUG\)](#)
- [Bromothymol blue \(GoldBio Catalog # B-750\)](#)
- [Deoxycholic acid sodium salt \(GoldBio Catalog # D-070\)](#)

References

Deisingh, A. and Thompson, M. (2004). Strategies for the detection of *Escherichia coli* O157:H7 in foods. *Journal of Applied Microbiology*, 96(3), 419-429. Doi:10.1111/j.1365-2672.2003.02170.x.

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