DYS SEE

M9 medium.

Protocols

M9 Medium

M9 is a minimal medium for bacteria. It is used as an alternative to LB for many purposes, mainly because it does emit fluorescence and is transparent. However, cells grow somewhat slower than in a rich medium. This is a modified protocol from Sambrook's Molecular Cloning (2001).

Procedure

Preparing the 5X stock:

Add the following reagents to a 2-liter flask:

- 64 g Na_2HPO_4, seven hydrate
- 15 g KH_2PO_4
- 5 g NH_4Cl
- 2.5 g NaCl
- 1 liter of high-quality distilled water

Once the ingredients are added, heat with stirring until the components are completely dissolved. Pour the solution into smaller bottles with loosened caps and the autoclave at 15 lb/in2 for 15 min. If you wish to add antibiotics or nutritional supplements, do this only after the autoclave cycle is complete, as the high temperature may destroy these components. Wait until the bottle is less than 50°C (it should be warm to touch), and then add the components. After the bottles cool to below 40°C, the caps can be tightened and the concentrated medium stored indefinitely at room temperature.

Preparing the 1X Working Solution:

To make 1X working solution, the 5X media should be diluted to 1× with high quality sterile distilled water.

Add the following sterile solutions for 1 liter of medium

- 1 ml 1 M $MgSO_4.7H_2O$
- 10 ml 20% D-glucose

Typically, several additional components are also added to make a complete medium.

- 34 ml 0.5% vitamin B1 (thiamine)
- 10 ml 20% Casamino Acids
- 5 µl 1M $CaCl_2$
- Antibiotic for selection

References

- Formulation sourced from an article by Karen Elbing and Roger Brent. Media Preparation and Bacteriological Tools. Current Protocols in Molecular Biology (2002) 1.1.1-1.1.7.
- Sambrook & Russel, Molecular Cloning (2001) 3rd edition

Protocols