



Standard Laboratory Operating Procedure #102 Media Preparation for IVC Cultures

Laboratory: Biotechnology

Location: Ag Biotech Academy

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General: The goal of this procedure is to prepare media containing antimicrobials and fungicides for controlling contamination in IVC cultures.

Safety: Safety Glasses

Materials:

Murashige & Skoog Basal Media with sucrose & Gelzan (www.phytotechlab.com)

Fungicide (purchased at your local garden center)

Antibiotic-Antimycotic Solution (www.sigmaaldrich.com)

1 dram vials (Carolina Biological) containing the following media and labels:

Vial #1: Medium only

Vial #2: Medium with fungicide

Vial #3: Medium with antimicrobial agent

Vial #4: Medium with fungicide and antimicrobial agent

Procedure:

To prepare Vial #1 medium:

1. Weigh out 1.8 g of Murashige & Skoog Basal Media.
2. Add the Murashige & Skoog Basal Media to 50 ml dH₂O in a 150 ml Erlenmeyer beaker.
3. Heat to boiling.
4. Allow to cool to approximately 60⁰ C.
5. For Vial #1, pipette 2 ml into the vials labeled 1 using a sterile pipette.

To prepare Vial #2 medium:

6. Repeat steps 1-4.
7. Add 5 ml of the fungicide to the Murashige & Skoog Basal Media.
8. Pipette 2 ml into the vials labeled 2 using a sterile pipette.

To prepare Vial #3 medium:

9. Repeat steps 1-4.
10. Add 13 μ l of the Antibiotic-Antimycotic Solution to the Murashige & Skoog Basal Media.
11. Pipette 2 ml into the vials labeled 3 using a sterile pipette.

To prepare Vial #4 medium:

12. Repeat steps 1-4.
13. Add 5 ml of the fungicide AND 13 μ l of the Antibiotic-Antimycotic Solution to the Murashige & Skoog Basal Media.
14. Pipette 2 ml into the vials labeled 4 using a sterile pipette.