

Fusarium graminearum Media Recipes

Lactic acid PDA (APDA)

*Used for isolation and for inoculating corn and MB broth.
Lactic acid inhibits bacterial growth.*

1. Add 39 g PDA (Fisher #DF0013176) to 1 L distilled H₂O.
2. Autoclave for 20 minutes on the fluid setting.
3. Cool in a water bath to ~50°C.
4. Add 1 mL 85% Lactic acid and swirl to mix.
5. Pour ~30 mL per plate.

Carnation Leaf Agar (CLA)

Used for getting good macroconidia formation for identification.

1. Collect carnation leaves and cut into 3-5 mm² pieces.
2. Microwave ~30 seconds to sterilize and dry.
3. Make 2% water
 - a. Add 20 g agar (Fisher # BP1423-500) to 1 L of H₂O.
 - b. Autoclave for 20 minutes on the fluid setting.
 - c. Cool in a water bath to ~50°C.
4. Add 10-15 pieces of carnation to each plate.
5. Pour ~30 mL water agar into each plate.

Mung Bean Broth (MBB)

Used for spore production in liquid culture.

1. Bring 1 L distilled H₂O to a boil.
2. Remove from heat and allow to sit 30 seconds.
3. Add 40 g of mung beans and allow to steep 10 minutes.
4. Fix a Buchner funnel to a 2 L flask and cover the holes with cheesecloth. Filter the beans from the broth.
5. Divide the broth as needed into flasks and put a sponge cork in the top.
6. Autoclave 20 minutes on the fluid setting.
7. Allow broth to cool overnight before inoculating.

Mung Bean Agar (MBA)

Used for spore production on culture plates. Protocol developed by the lab of Dr. Ruth Dill-Macky.

1. **Boil water** in 2 L beaker.
2. **Place** beans in water and boil for **23 min**.
NOTE: Time from when the water has returned to the boil. The beans will have started to crack open by 23 min (this may happen as early as 15 min). Good spore production is dependent on a cooking time of 23 minutes.
3. **Remove** bean broth from heat.
4. **Mark** 1 liter volume on the side of a 2-liter Erlenmeyer flask.
5. **Place** a large **Buchner funnel** on the flask.
6. **Line** the **funnel** using two **gauze pads** (4-6 layers of cheesecloth may be substituted). Ensure that the gauze/cheesecloth covers the funnel holes.
7. **Pour** the bean broth slowly into the flask through the funnel
8. **Discard the beans** (and gauze) from the funnel
9. **Soak** the **funnel and beaker** in water
NOTE: The residue is difficult to remove once dried onto surfaces!
10. **Adjust** the total **volume** of broth up to 1 liter by adding **distilled water** to the mark on the flask
11. **Add** 15 grams of **agar (Fisher # BP1423-500)** and **mix**.
12. **Autoclave** at 115 °C for 20 minutes, fluid setting
13. **Cool in a water bath to ~50°C**.
14. **Pour ~30 mL per plate**.