



## Grape Agar Powder Premix

### Instructions for Use:

**For best results and maximum yield, use a microwave. Please note packets are for single use only; partial use will adversely affect ingredient ratios.**

**Important Note:** Never pour H<sub>2</sub>O onto the grape agar powder premix. Pouring H<sub>2</sub>O onto the mix can cause excessive clumping to occur.

### Microwave Instructions:

1. Add 500ml room temperature dH<sub>2</sub>O to a 1L glass flask or beaker.
2. Slowly pour packet contents from 1 packet onto the dH<sub>2</sub>O in the flask while stirring gently until all powdered components have dissolved leaving no clumps.
3. Microwave on high until the mixture begins to vigorously boil and climb up the sides of the flask (~4-5 min).
  - a. **DO NOT add stir bar to the flask before microwaving.**
4. Upon boiling, remove the flask from the microwave (**Warning: Container will be extremely hot!**) and gently stir with a stir plate for 5 to 10 minutes or until the solution cools to approximately 70-75°C. May also stir solution by hand to achieve the same results.
5. Microwave again on high until the mixture begins to boil and climb up the sides of the flask (~2-3min).
  - a. **DO NOT add stir bar to the flask before microwaving.**
6. Upon boiling, remove the flask from the microwave (**Warning: Container will be extremely hot!**) and gently stir with a stir plate (tilt flask and let stir bar run down side of flask) for at least 15 minutes or until the solution cools to approximately 55-60°C.
7. Pour plates.

### Helpful Hints:

Place the glass flask or beaker into a secondary container to capture any overflow that may occur during boiling. Spillage can be prevented by keeping a close eye on the mix while microwaving and removing once the mixture begins to boil and climb up the sides of the flask. Cook time will be affected by model, age and power of your microwave.

### Hot Plate-Stirrer Instructions:

1. Add 500ml room temperature dH<sub>2</sub>O to a 1L glass flask or beaker.
2. Add stir bar to the glass flask or beaker (Tilt flask and let stir bar run down the side of the flask).
3. Start the stirrer on medium according to the manufacturer's instructions.
4. Slowly pour contents from 1 packet onto the dH<sub>2</sub>O in the flask.
5. Turn on heat and slowly heat to a vigorous boil. Let boil until the grape agar solution begins to climb up the sides of the flask.
6. Once bubbles are observed, turn off the heat and continue to stir gently with the stir plate for 5 – 10 minutes while the solution cools to approximately 70-75°C.
7. Turn on heat again and slowly heat to a boil. Let boil until the grape agar solution begins to climb up the sides of the flask.
8. Once bubbles are observed, turn off the heat and continue to stir gently with the stir plate for 5 – 10 minutes while the solution cools to approximately 60 °C.
9. Pour plates.

### Autoclave Instructions:

1. Add 500ml room temperature dH<sub>2</sub>O to a 1L glass flask or beaker.
2. Slowly pour contents from 1 packet onto the dH<sub>2</sub>O in the flask while stirring gently until all powdered components have dissolved leaving no clumps.
3. **DO NOT add stir bar to the flask before autoclaving.**
4. For 500ml of solution, autoclave on the **liquid cycle** for 10 minutes. For 1000ml of solution autoclave for 15 minutes.
  - a. Use only borosilicate (such as Pyrex) flasks with vented closures.
5. Make sure seals on containers of liquids are loose so vapor expanding during heating will not cause an explosion.
6. Remove glass flask or beaker from autoclave (**Warning: Container will be extremely hot!**) and gently stir with a stir plate for at least 15 minutes or until the solution cools to approximately 55-60°C.
7. Pour plates.

### Troubleshooting:

1. Uneven agar surface due to bubbles. **Solution:** Use a flame on the agar surface for a smooth, glassy surface.
2. Agar does not fully solidify. **Solution:** Microwave or reheat the agar and bring the agar to a boil for a third time.
3. Agar still does not fully solidify after boiling a third time. **Solution:** If the agar does not solidify after a third boiling, it is probably due to environmental factors (humidity), and therefore will need slightly more agar. Please contact Genesee Scientific at 800.789.5550 or [support@geneseesci.com](mailto:support@geneseesci.com). We will send a small supplemental bottle of Grape Agar Powder Premix along with a replacement for any wasted Grape Agar Powder Premix.