



## Pseudomonas isolation agar

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### ABSTRACT

This protocol describes how to make 1 L of *Pseudomonas* isolation agar for the culturing of *Pseudomonas* bacterial species on plates.

The use of a genus specific isolation media such as *Pseudomonas* isolation media reduces the risk of plate contamination with a different bacterial species when trying to isolate a particular bacterial strain.

### PROTOCOL STATUS

#### Working

We use this protocol in our group and it is working

### GUIDELINES

Once *Pseudomonas* isolation media has been autoclaved, all work should be carried out in a laminar flow hood to prevent contamination.

1 L of *Pseudomonas* isolation agar makes ~20 plates.

At 4 °C unused plates can be stored for around 2 weeks prior to use.

Once used, plates containing bacterial colonies can be stored in air-tight container/bag at 4 °C for a maximum of 2 weeks.

Prior to disposal, plates containing bacterial colonies must be wrapped in Parafilm and autoclaved.

### MATERIALS

| NAME ▾                     | CATALOG # ▾ | VENDOR ▾                           |
|----------------------------|-------------|------------------------------------|
| Glycerol A.R               | CHE2068     | Scientific Laboratory Supplies Ltd |
| Pseudomonas Isolation agar | 17208-500g  | Sigma Aldrich                      |

### SAFETY WARNINGS

Read SDS for *Pseudomonas* Isolation Agar before use.

Bottle can be extremely hot immediately following autoclave. Use thermal gloves for handling.

- 1 Add 45.03 g *Pseudomonas* isolation agar and 20 mL of 100% glycerol per 980 mL of water.
- 2 Mix well by inverting bottle several times until powder dissolved.
- 3 Sterilize by autoclaving for 20 min at 15 psi (1.05 kg/cm<sup>2</sup>) on liquid cycle.
- 4 Following autoclave (while media still liquid) pour *Pseudomonas* isolation agar from bottle into plates within a laminar flow hood. Allow ~50 mL per plate.

- 5 Leave plates in laminar hood until agar sets.
- 6 When agar has set, invert plates and store in an air tight bag at 4 °C.
- 7 Plates should be removed from 4 °C at least 1 h before use and placed at room temperature. To prevent condensation from lid dropping onto agar surface, wipe lids dry with Kimwipe before placing at room temperature and store plates inverted.



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