

## 457: MINERAL MEDIUM (BRUNNER)

This recipe contains strain-specific modifications for *Acinetobacter sp.* DSM 7324 \*

Final pH: 6.9

Final volume: 1000 ml

|   |         |    |
|---|---------|----|
| Na <sub>2</sub> HPO <sub>4</sub>                | 2.44    | g  |
| KH <sub>2</sub> PO <sub>4</sub>                 | 1.52    | g  |
| (NH <sub>4</sub> ) <sub>2</sub> SO <sub>4</sub> | 0.50    | g  |
| MgSO <sub>4</sub> x 7 H <sub>2</sub> O          | 0.20    | g  |
| CaCl <sub>2</sub> x 2 H <sub>2</sub> O          | 0.05    | g  |
| <b>Trace element solution SL-4</b>              | 10.00   | ml |
| Acetanilide                                     | 5.00    | mM |
| Distilled water                                 | 1000.00 | ml |

1. Adjust pH to 6.9.
2. Prepare a separate solution of the phosphates and autoclave separately. Combine the two solutions after cooling.
3. Rehydrate and cultivate lyophilized cells in complex medium (e.g. medium 1, 220 or 535). After this reactivation, cultivate in mineral medium 457 with the appropriate carbon source.

\* add 5 mmol/l acetanilide from a filter sterilized stock solution (30 mmol/l)

### Trace element solution SL-4

|  |        |    |
|--|--------|----|
| EDTA                                   | 0.50   | g  |
| FeSO <sub>4</sub> x 7 H <sub>2</sub> O | 0.20   | g  |
| <b>Trace element solution SL-6</b>     | 100.00 | ml |
| Distilled water                        | 900.00 | ml |

### Trace element solution SL-6 (from medium 27)

|   |         |    |
|---|---------|----|
| ZnSO <sub>4</sub> x 7 H <sub>2</sub> O                | 0.10    | g  |
| MnCl <sub>2</sub> x 4 H <sub>2</sub> O                | 0.03    | g  |
| H <sub>3</sub> BO <sub>3</sub>                        | 0.30    | g  |
| CoCl <sub>2</sub> x 6 H <sub>2</sub> O                | 0.20    | g  |
| CuCl <sub>2</sub> x 2 H <sub>2</sub> O                | 0.01    | g  |
| NiCl <sub>2</sub> x 6 H <sub>2</sub> O                | 0.02    | g  |
| Na <sub>2</sub> MoO <sub>4</sub> x 2 H <sub>2</sub> O | 0.03    | g  |
| Distilled water                                       | 1000.00 | ml |

## 457: MINERAL MEDIUM (BRUNNER)

### Main sol. 1 (from medium 1)

|                        |         |    |
|------------------------|---------|----|
| Peptone                | 5.00    | g  |
| Meat extract           | 3.00    | g  |
| Agar, for solid medium | 15.00   | g  |
| Distilled water        | 1000.00 | ml |

1. Adjust pH to 7.0.
2. For Bacillus strains the addition of 10.0 mg  $\text{MnSO}_4 \times \text{H}_2\text{O}$  is recommended for sporulation.

### Main sol. 220 (from medium 220)

|                 |         |    |
|-----------------|---------|----|
| Casein peptone  | 15.00   | g  |
| Soy peptone     | 5.00    | g  |
| NaCl            | 5.00    | g  |
| Agar            | 15.00   | g  |
| Distilled water | 1000.00 | ml |

Adjust pH to 7.3. Medium is identical with Tryptone Soya Agar (Oxoid CM 131).

### Main sol. 535 (from medium 535)

|  |         |    |
|--|---------|----|
| Trypticase soy broth (BBL 11768, Oxoid CM129 or Merck <del>30469</del> ) | 30.00   | g  |
| Agar   | 20.00   | g  |
| Distilled water  | 1000.00 | ml |

1. pH 7.3
2. Autoclave at 121°C for 15 min.