

457: MINERAL MEDIUM (BRUNNER)

This recipe contains strain-specific modifications for *Pseudomonas putida* DSM 6755 *

Final pH: * 7.5

Final volume: 1000 ml

Na ₂ HPO ₄	2.44	g
KH ₂ PO ₄	1.52	g
(NH ₄) ₂ SO ₄	0.50	g
MgSO ₄ x 7 H ₂ O	0.20	g
CaCl ₂ x 2 H ₂ O	0.05	g
Trace element solution SL-4	10.00	ml
Yeast extract	100.00	mg/l
Sodium chloroacetate	20.00	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.

* PH 7.5, with 100 mg/l yeast extract and 20 mM sodium chloroacetate from a filter-sterilized 1 mol/l stock solution

Trace element solution SL-4

EDTA	0.50	g
FeSO ₄ x 7 H ₂ O	0.20	g
Trace element solution SL-6	100.00	ml
Distilled water	900.00	ml

Trace element solution SL-6 (from medium 27)

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

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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 30469)	30.00	g
Agar	20.00	g
Distilled water	1000.00	ml

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