

202. DESULFONEMA MAGNUM MEDIUM

Solution A:

NaCl	25.00	g
MgCl ₂ x 6 H ₂ O	5.60	g
MgSO ₄ x 7 H ₂ O	6.80	g
CaCl ₂ x 2 H ₂ O	1.40	g
KCl	0.72	g
KH ₂ PO ₄	0.14	g
NH ₄ Cl	0.25	g
Trace element solution SL-10 (see medium 320)	1.00	ml
Selenite-tungstate solution (see medium 385)	1.00	ml
Na-resazurin solution (0.1% w/v)	0.50	ml
Distilled water	1000.00	ml

Solution B:

Na-benzoate	0.60	g
Distilled water	10.00	ml

Solution C:

Vitamin solution (see medium 141)	10.00	ml
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Solution D:

KAl(SO ₄) ₂ x 12 H ₂ O	0.48	g
Distilled water	10.00	ml

Solution E:

Na ₂ CO ₃	1.00	g
Distilled water	20.00	ml

Solution F:

Na ₂ S x 9 H ₂ O	0.30	g
Distilled water	10.00	ml

Solution A is sparged with 80% N₂ and 20% CO₂ gas mixture for 30 – 45 min to reach a pH of around 6, then distributed under the same gas atmosphere into anoxic Hungate-type tubes and autoclaved. *Solutions B, D, and F* are autoclaved separately under 100% N₂ gas. *Solution E* is autoclaved under 80% N₂ and 20% CO₂ gas mixture. *Solution C* is prepared under 100% N₂ gas and sterilized by filtration.

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To complete the medium appropriate amounts of *solutions B to F* are added to the sterile *solution A* in the sequence as indicated. Final pH of the medium should be 7.0. After completion the medium should equilibrate overnight and a white precipitate should be apparent.

Note: Addition of 10 - 20 mg sodium dithionite per liter (e.g. from 5% (w/v) solution freshly prepared under N₂ and filter-sterilized) just before inoculation may stimulate growth at the beginning. For transfers use 5 - 10% inoculum.