

346. DESULFALLAS MEDIUM

Solution A:

Na ₂ SO ₄	3.00	g
KH ₂ PO ₄	0.20	g
NH ₄ Cl	0.30	g
CaCl ₂ x 2 H ₂ O	0.15	g
MgCl ₂ x 6 H ₂ O	0.40	g
NaCl	1.00	g
Trace element solution SL-10 (see medium 320)	1.00	ml
Na-resazurin solution (0.1% w/v)	0.50	ml
Distilled water	965.00	ml

Solution B:

NaHCO ₃	1.00	g
Distilled water	20.00	ml

Solution C:

Ethanol	1.00	ml
Distilled water	10.00	ml

Solution D:

Na-benzoate	0.70	g
Distilled water	10.00	ml

Solution E:

Rumen fluid, clarified (see medium 1310)	1.00	ml
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Solution F:

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

Na-dithionite	25.00	mg
Distilled water	1.00	ml

Solution H:

Na ₂ S x 9 H ₂ O	50.00	mg
Distilled water	1.70	ml

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Sparge *solution A* with 100% N₂ gas for 30 - 45 min to make it anoxic. Thereafter, dispense under same gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. *Solution B* is autoclaved separately under 80% N₂ and 20% CO₂ gas atmosphere. Anoxic stock *solutions C, D, E, F, G* and *H* are prepared separately under 100% N₂ gas atmosphere. Filter sterilize *solutions D, F* and *G*. To complete the medium appropriate amounts of *solutions B* to *H* are added to the sterile *solution A* in the sequence as indicated. Adjust pH of the complete medium to 7.2 - 7.5, if necessary.