

792d: MUCITHERMUS MEDIUM

Solution A	949.00	ml
Solution B	20.00	ml
Solution C	10.00	ml
Solution D	10.00	ml
Solution E	10.00	ml
Solution F	10.00	ml

1. Sparge solution A with 80% N₂ and 20% CO₂ gas mixture for 30 - 45 min to make it anoxic, then 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 mixture and solutions C, D, E and F are autoclaved under 100% N₂ gas. To complete the medium appropriate amounts of solutions B, C, D, E and F are added to solution A. Adjust pH of the complete medium to 6.5 - 7.0. After inoculation pressurize vials to 1 bar overpressure with sterile 80% N₂ and 20% CO₂ gas mixture.

2. Note: Solution A has to be prepared freshly. It cannot be stored for a long period of time.

Solution A

NaCl	13.85	g
MgSO ₄ × 7 H ₂ O	3.50	g
MgCl ₂ × 6 H ₂ O	2.75	g
CaCl ₂ × 2 H ₂ O	0.38	g
KCl	0.33	g
NH ₄ Cl	0.50	g
NaBr	0.05	g
H ₃ BO ₃	15.00	mg
SrCl ₂ × 6 H ₂ O (0.1% w/v)	7.00	ml
KI (0.01% w/v)	0.50	ml
Wolfe's mineral elixir	1.00	ml
Sodium resazurin (0.1% w/v)	0.50	ml
Distilled water	940.00	ml

Solution B

Na ₂ CO ₃	1.00	g
Distilled water	20.00	ml

Solution C

KH ₂ PO ₄	0.50	g
Distilled water	10.00	ml

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Solution D

Trypticase peptone	1.00	g
Distilled water	10.00	ml

Solution E

Yeast extract	1.00	g
Distilled water	10.00	ml

Solution F

$\text{Na}_2\text{S} \times 9 \text{H}_2\text{O}$	0.30	g
Distilled water	10.00	ml

Wolfe's mineral elixir (from medium 792)

$\text{MgSO}_4 \times 7 \text{H}_2\text{O}$	30.00	g
$\text{MnSO}_4 \times \text{H}_2\text{O}$	5.00	g
NaCl	10.00	g
$\text{FeSO}_4 \times 7 \text{H}_2\text{O}$	1.00	g
$\text{CoCl}_2 \times 6 \text{H}_2\text{O}$	1.80	g
$\text{CaCl}_2 \times 2 \text{H}_2\text{O}$	1.00	g
$\text{ZnSO}_4 \times 7 \text{H}_2\text{O}$	1.80	g
$\text{CuSO}_4 \times 5 \text{H}_2\text{O}$	0.10	g
$\text{AlK}(\text{SO}_4)_2 \times 12 \text{H}_2\text{O}$	0.18	g
H_3BO_3	0.10	g
$\text{Na}_2\text{MoO}_4 \times 2 \text{H}_2\text{O}$	0.10	g
$(\text{NH}_4)_2\text{Ni}(\text{SO}_4)_2 \times 6 \text{H}_2\text{O}$	2.80	g
$\text{Na}_2\text{WO}_4 \times 2 \text{H}_2\text{O}$	0.10	g
Na_2SeO_4	0.10	g
Distilled water	1000.00	ml

First adjust pH to 1.0 with diluted H_2SO_4 , then add and dissolve the salts.