

792. PYROLOBUS FUMARI I MEDIUM

Solution A:

NaCl	13.85	g
MgSO ₄ x 7 H ₂ O	3.50	g
MgCl ₂ x 6 H ₂ O	2.75	g
CaCl ₂ x 2 H ₂ O	0.38	g
KCl	0.33	g
NaBr	0.05	g
H ₃ BO ₃	15.00	mg
SrCl ₂ x 6 H ₂ O solution (0.1% w/v)	7.00	ml
KI solution (0.01% w/v)	0.50	ml
Wolfe's mineral elixir (see next page)	1.00	ml
Na-resazurin solution (0.1% w/v)	0.50	ml
Double distilled water	980.00	ml

Solution B:

KNO ₃	1.00	g
KH ₂ PO ₄	0.50	g
Distilled water	10.00	ml

Solution C:

Seven vitamins solution (see medium 503)	1.00	ml
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Solution D:

Na ₂ S x 9 H ₂ O	0.15	g
Distilled water	10.00	ml

Sparge *solution A* with 80% H₂ and 20% CO₂ gas mixture for 30 - 45 min to make it anoxic, then dispense under same gas atmosphere into anoxic serum vials (e.g., 20 ml medium in 100 ml serum bottles) and autoclave. *Solutions B* and *D* are autoclaved separately under 100% N₂ gas atmosphere and *solution C* is prepared under 100% N₂ gas and sterilized by filtration. To complete the medium appropriate amounts of *solutions B*, *C* and *D* are added to *solution A*. Adjust pH of the complete medium to 5.8 using a sterile anoxic stock solution of 5% (w/v) Na₂CO₃, if necessary. After inoculation pressurize vials to 1 bar overpressure with sterile 80% H₂ and 20% CO₂ gas mixture. *Note: Solution A has to be prepared freshly. It cannot be stored for a long period of time.*

Continued on next page.

Wolfe's mineral elixir:

MgSO ₄ x 7 H ₂ O	30.00	g
MnSO ₄ x H ₂ O	5.00	g
NaCl	10.00	g
FeSO ₄ x 7 H ₂ O	1.00	g
CoCl ₂ x 6 H ₂ O	1.80	g
CaCl ₂ x 2 H ₂ O	1.00	g
ZnSO ₄ x 7 H ₂ O	1.80	g
CuSO ₄ x 5 H ₂ O	0.10	g
KAl(SO ₄) ₂ x 12 H ₂ O	0.18	g
H ₃ BO ₃	0.10	g
Na ₂ MoO ₄ x 2 H ₂ O	0.10	g
(NH ₄) ₂ Ni(SO ₄) ₂ x 6 H ₂ O	2.80	g
Na ₂ WO ₄ x 2 H ₂ O	0.10	g
Na ₂ SeO ₄	0.10	g
Distilled water	1000.00	ml

First adjust pH to 1.0 with diluted H₂SO₄, then add and dissolve the salts.