

1020. SULFURICURVUM (MBM) MEDIUM

NaNO ₃	0.2	g
KH ₂ PO ₄	0.2	g
NH ₄ Cl	0.2	g
MgCl ₂ x 6 H ₂ O	0.4	g
KCl	0.2	g
CaCl ₂ x 2 H ₂ O	0.1	g
Na-resazurin solution (0.1% w/v)	0.5	ml
Trace element solution SL-4 (see medium 14)	2.0	ml
Na ₂ S ₂ O ₃ x 5 H ₂ O	2.5	g
Distilled water	1000.0	ml

Dissolve ingredients, except SL-4 trace element solution and sodium thiosulfate, then sparge solution with 80% N₂ and 20% CO₂ gas mixture for 30 – 45 min to make it anoxic. Adjust pH to 7.0, then distribute medium under 80% N₂ and 20% CO₂ gas atmosphere into anoxic Hungate-type tubes or serum vials (e.g., 20 ml medium in 100 ml bottles) and autoclave. Prior to inoculation, add trace elements and thiosulfate from anoxic stock solutions prepared under 100% N₂ gas and sterilized by filtration. Adjust pH of complete medium to 7.0, if necessary. After inoculation, pressurize culture vessels to 0.5 bar overpressure with sterile 100% H₂ gas.