

## 1020: SULFURICURVUM (MBM) MEDIUM

NaNO <sub>3</sub>	0.20	g
KH <sub>2</sub> PO <sub>4</sub>	0.20	g
NH <sub>4</sub> Cl	0.20	g
MgCl <sub>2</sub> x 6 H <sub>2</sub> O	0.40	g
KCl	0.20	g
CaCl <sub>2</sub> x 2 H <sub>2</sub> O	0.10	g
<b>Trace element solution SL-4</b>	2.00	ml
Na <sub>2</sub> S <sub>2</sub> O <sub>3</sub> x 5 H <sub>2</sub> O	2.50	g
Distilled water	1000.00	ml

1. Dissolve ingredients, except sodium thiosulfate, then sparge solution with 80% N<sub>2</sub> and 20% CO<sub>2</sub> gas mixture for 30 - 45 min to make it anoxic. Adjust pH to 7.0, then distribute medium under 80% N<sub>2</sub> and 20% CO<sub>2</sub> 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 thiosulfate from an anoxic stock solution prepared under 100% N<sub>2</sub> gas and sterilized by filtration. Adjust pH of complete medium to 7.0, if necessary.
2. After inoculation, pressurize culture vessels to 0.5 bar overpressure with sterile 100% H<sub>2</sub> gas.

### Trace element solution SL-4 (from medium 14)

Na <sub>2</sub> -EDTA	0.50	g
FeSO <sub>4</sub> x 7 H <sub>2</sub> O	0.20	g
ZnSO <sub>4</sub> x 7 H <sub>2</sub> O	0.10	g
MnCl <sub>2</sub> x 4 H <sub>2</sub> O	0.03	g
H <sub>3</sub> BO <sub>3</sub>	0.30	g
CoCl <sub>2</sub> x 6 H <sub>2</sub> O	0.20	g
CuCl <sub>2</sub> x 2 H <sub>2</sub> O	0.01	g
NiCl <sub>2</sub> x 6 H <sub>2</sub> O	0.02	g
Na <sub>2</sub> MoO <sub>4</sub> x 2 H <sub>2</sub> O	0.03	g
Distilled water	1000.00	ml

First dissolve EDTA in distilled water and adjust pH to 7.0 using 2 N NaOH; then add other compounds.