

**122. CLOSTRIDIUM THERMOCELLUM MEDIUM**

(NH <sub>4</sub> ) <sub>2</sub> SO <sub>4</sub>	1.30	g
MgCl <sub>2</sub> x 6 H <sub>2</sub> O	2.60	g
KH <sub>2</sub> PO <sub>4</sub>	1.43	g
K <sub>2</sub> HPO <sub>4</sub>	5.50	g
CaCl <sub>2</sub> x 2 H <sub>2</sub> O	0.13	g
Na <sub>2</sub> -β-glycerol phosphate x 4 H <sub>2</sub> O	6.00	g
FeSO <sub>4</sub> x 7 H <sub>2</sub> O solution (0.1% w/v in 0.1 N H <sub>2</sub> SO <sub>4</sub> )	1.10	ml
L-Glutathione reduced	0.25	g
Yeast extract	4.50	g
Na-resazurin solution (0.1% w/v)	0.50	ml
Cellobiose	5.00	g
Distilled water	1000.00	ml

Dissolve ingredients except cellobiose, sparge medium with 80% N<sub>2</sub> and 20% CO<sub>2</sub> gas mixture for 30 – 45 min to make it anoxic. Then adjust pH to 7.0 - 7.2, distribute under same gas atmosphere in anoxic Hungate-type tubes or serum vials and autoclave.

Cellobiose is added to the sterile medium from an anoxic 10% (w/v) stock solution prepared under 100% N<sub>2</sub> gas and sterilized by filtration. Some strains can be adapted to cellulose as substrate using 10.00 g/l cellulose (Avicel or MN 301, MACHEREY-NAGEL).

*Note: A white precipitate forms after mixing the ingredients of this medium, but this has no negative effect on growth.*