

**539. RUMINICLOSTRIDIUM CELLOBIOPARUM MEDIUM**

KH <sub>2</sub> PO <sub>4</sub>	0.20	g
NH <sub>4</sub> Cl	0.30	g
NaCl	1.00	g
MgCl <sub>2</sub> x 6 H <sub>2</sub> O	0.40	g
KCl	0.50	g
CaCl <sub>2</sub> x 2 H <sub>2</sub> O	0.15	g
Trace element solution SL-10 (see medium 320)	1.00	ml
Yeast extract	0.50	g
Na-resazurin solution (0.1% w/v)	0.50	ml
NaHCO <sub>3</sub>	4.50	g
Cellobiose	5.00	g
Na <sub>2</sub> S x 9 H <sub>2</sub> O	0.40	g
Distilled water	1000.00	ml

Dissolve ingredients (except bicarbonate, cellobiose and sulfide), then sparge medium with 80% N<sub>2</sub> and 20% CO<sub>2</sub> gas mixture for 30 – 45 min to make it anoxic. Dispense medium under same gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. After sterilization add cellobiose (sterilized by filtration) and sulfide from sterile anoxic stock solutions prepared under 100% N<sub>2</sub> gas and bicarbonate from a sterile anoxic stock solution prepared under 80% N<sub>2</sub> and 20% CO<sub>2</sub> gas mixture. Adjust pH of complete medium to 7.0, if necessary.

*Note: Some strains can be adapted to cellulose as substrate using 5.00 g/l cellulose powder MN 301 (MACHEREY-NAGEL).*