## **Microorganisms**



## 1233: CALDICOPROBACTER MEDIUM

NaCl	0.50	
IVaCi	0.50	g
KCI	0.20	g
MgCl <sub>2</sub> x 6 H <sub>2</sub> O	0.18	g
NH <sub>4</sub> Cl	0.10	g
HEPES (SIGMA)	4.80	g
$Na_2HPO_4 \times 7 H_2O$	54.00	mg
$Fe(NH_4)_2(SO_4)_2 \times 6 H_2O (0.1\% \text{ w/v})$	4.00	ml
$CaCl_2 \times 2 H_2O (0.1\% w/v)$	2.00	ml
$MnCl_2 \times 4 H_2O (0.1\% \text{ w/v})$	1.00	ml
Yeast extract (OXOID)	5.00	g
Tryptone (BD Bacto)	2.50	g
Sodium resazurin (0.1% w/v)	0.50	ml
L-Cysteine HCl x H <sub>2</sub> O	1.00	g
D-Glucose	5.00	g
Distilled water	1000.00	ml

- 1. Dissolve ingredients (except cysteine and glucose), then sparge medium with 100%  $N_2$  gas for 30 45 min to make it anoxic. Add solid cysteine and adjust pH to 7.3 7.5, then dispense medium under 100%  $N_2$  gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. Before inoculation, add glucose from a sterile anoxic stock solution prepared under 100%  $N_2$  gas.
- 2. Addition of 10 20 mg sodium dithionite per liter (e.g. from 5% (w/v) solution, freshly prepared under  $N_2$  and filter-sterilized) can be used to completely reduce the medium and may stimulate growth at the beginning.