

**985. TEPIDIBACTER MEDIUM**

NaCl	18.00	g
MgCl <sub>2</sub> x 6 H <sub>2</sub> O	4.00	g
KCl	0.34	g
NH <sub>4</sub> Cl	0.25	g
CaCl <sub>2</sub>	0.11	g
K <sub>2</sub> HPO <sub>4</sub>	0.18	g
Fe(NH <sub>4</sub> ) <sub>2</sub> (SO <sub>4</sub> ) <sub>2</sub> x 7 H <sub>2</sub> O	20.00	mg
Trace element solution SL-10 (see medium 320)	1.00	ml
Selenite-tungstate solution (see medium 385)	1.00	ml
Yeast extract (OXOID)	0.20	g
Proteose peptone (BD Difco)	10.00	g
Na-resazurin solution (0.1% w/v)	0.50	ml
NaHCO <sub>3</sub>	5.00	g
Vitamin solution (see medium 141)	10.00	ml
Na <sub>2</sub> S x 9 H <sub>2</sub> O	0.50	g
Distilled water	1000.00	ml

Dissolve ingredients (except bicarbonate, vitamins 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. Add vitamins (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 atmosphere. The pH of the complete medium should be 6.5 - 7.0.