

**1051. TEPIDANAEROBACTER MEDIUM**

KH <sub>2</sub> PO <sub>4</sub>	0.14	g
MgCl <sub>2</sub> x 6 H <sub>2</sub> O	0.20	g
CaCl <sub>2</sub> x 2 H <sub>2</sub> O	0.15	g
NH <sub>4</sub> Cl	0.54	g
Trace element solution (see medium 144)	1.00	ml
Na-resazurin solution (0.1% w/v)	0.50	ml
Na <sub>2</sub> CO <sub>3</sub>	1.50	g
Yeast extract	2.30	g
Vitamin solution (see medium 141)	2.00	ml
D-Glucose	2.20	g
L-Cysteine-HCl x H <sub>2</sub> O	0.30	g
Na <sub>2</sub> S x 9 H <sub>2</sub> O	0.30	g
Distilled water	1000.00	ml

Dissolve ingredients (except carbonate, yeast extract, vitamins, glucose, cysteine 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 yeast extract, vitamins (sterilized by filtration), glucose, cysteine and sulfide from sterile anoxic stock solutions prepared under 100% N<sub>2</sub> gas and carbonate from a sterile anoxic stock solution prepared under 80% N<sub>2</sub> and 20% CO<sub>2</sub> gas mixture. Adjust pH of the complete medium to 7.0-7.2, if necessary.

For DSM 21860 omit glucose, increase amount of ammonium chloride to 2.50 g/l, decrease amount of yeast extract to 0.20 g/l and supplement medium with 1.00 ml/l selenite-tungstate solution (see medium 385). As substrate add 1.10 g/l betaine to the autoclaved medium from an anoxic stock solution sterilized by filtration.