1533. MARINIFILUM MEDIUM (IRD MEDIUM)

KH$_2$PO$_4$ 0.3 g
K$_2$HPO$_4$ 0.3 g
NH$_4$Cl 1.0 g
NaCl 25.0 g
KCI 0.1 g
CaCl$_2$ x2H$_2$O 0.1 g
Cysteine HCl 0.5 g
Yeast extract Oxoid 1.0 g
Trace mineral element solution (see medium 941) 1 mL
Resazurin (0.1%) 1 mL
Distilled water 1000.00 mL

pH 7.0

Add after autoclave

MgCl$_2$ x 6 H$_2$O (150.0 g/l) 20 ml/l
NaHCO$_3$ (10%) 20 ml/l
Na$_2$S x 9 H$_2$O (2%) 20 ml/l
Glucose (1M) 20 ml/l

Prepare the medium without NaHCO$_3$, Na$_2$S x 9 H$_2$O, MgCl$_2$ x 6 H$_2$O and glucose. Boil the medium in a water bath and cool under a stream of N$_2$/CO$_2$ (80/20). Add the NaHCO$_3$ to the cooled medium (the pH should be 7.0) and dispense the medium into Hungate tubes or serum bottles under a stream of N$_2$/CO$_2$ (80/20). Seal the tubes under N$_2$/CO$_2$ (80/20) and autoclave. To the cooled medium at the appropriate amounts of Na$_2$S x 9 H$_2$O, MgCl$_2$ x 6 H$_2$O and glucose from sterile, anaerobic stock solutions.