483. DEFLUVIITALEA MEDIUM

KH$_2$PO$_4$ 0.3 g  
K$_2$HPO$_4$ 0.3 g  
NH$_4$Cl  1.0 g  
NaCl  1.0 g  
MgCl$_2$ x 6 H$_2$O  1.0 g  
CaCl$_2$ x 2 H$_2$O  0.1 g  
KCl  0.1 g  
Trace element solution (see medium 141) 10.0 ml  
Yeast extract  2.0 g  
Na-resazurin solution (0.1% w/v)  0.5 ml  
L-Cysteine-HCl x H$_2$O  0.5 g  
NaHCO$_3$  2.0 g  
D-Glucose  4.0 g  
Na$_2$S x 9 H$_2$O  0.3 g  
Distilled water  1000.0 ml  

Dissolve ingredients (except cysteine, bicarbonate, glucose and sulfide) and sparge medium with 80% N$_2$ and 20% CO$_2$ gas mixture for 30 – 45 min to make it anoxic. Add cysteine and adjust pH to 6.5, then dispense medium under same gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. After sterilization add glucose and sulfide from sterile anoxic stock solutions prepared under 100% N$_2$ gas and bicarbonate from a sterile anoxic stock solution prepared under 80% N$_2$ and 20% CO$_2$ gas mixture. Adjust pH of complete medium to pH 7.2 -7.4, if necessary.