1092. THERMOSULFIDIBACTER MEDIUM

Sea Salts (SIGMA) 30.0 g
Wolfe’s mineral elixier (see medium 792) 1.0 ml
Na-resazurin solution (0.1% w/v) 0.5 ml
Sulfur, powdered 3.0 g
NaHCO₃ 2.5 g
Yeast extract 2.0 g
Vitamin solution (see medium 141) 20.0 ml
Na₂S x 9 H₂O 0.5 g
Distilled water 1000.0 ml

Dissolve ingredients (except sulfur, bicarbonate, yeast extract, vitamins and sulfide) and
sparge medium with 80% H₂ and 20% CO₂ gas mixture for 30 - 45 min to make it
anoxic. Dispense medium under same gas atmosphere into anoxic serum vials (e.g.,
Balch-type tubes) containing already the appropriate amount of sulfur and autoclave at
110°C for 20 min. Add yeast extract, vitamins and sulfide form sterile anoxic stock
solutions prepared under 100% N₂ gas and bicarbonate from a sterile anoxic stock
solution prepared under 80% N₂ and 20% CO₂ gas mixture. The vitamin solution should
be sterilized by filtration. Adjust pH of the complete medium to 7.0-7.2. After inoculation
pressurize vessels with sterile 80% H₂ and 20% CO₂ gas mixture to 2 bar overpressure.