792a. THERMOVIBRIO AMMONIFICANS MEDIUM

Solution A:
- NaCl 20.00 g
- MgSO₄ x 7 H₂O 3.50 g
- MgCl₂ x 6 H₂O 2.75 g
- CaCl₂ x 2 H₂O 0.75 g
- KCl 0.33 g
- NaBr 0.05 g
- H₃BO₃ 15.00 mg
- SrCl₂ x 6 H₂O solution (0.1% w/v) 7.00 ml
- KI solution (0.01% w/v) 0.50 ml
- Wolfe's mineral elixir (see medium 792) 1.00 ml
- Na-resazurin solution (0.1% w/v) 0.50 ml
- Distilled water 980.00 ml

Solution B:
- KNO₃ 1.00 g
- KH₂PO₄ 0.50 g
- Distilled water 10.00 ml

Solution C:
- Na₂S x 9 H₂O 0.15 g
- Distilled water 10.00 ml

Sparge solution A with 80% H₂ and 20% CO₂ gas mixture for 30 - 45 min to make it anoxic, then dispense under same gas atmosphere into anoxic serum vials (e.g., 20 ml medium in 100 ml bottles) and autoclave. Solution B is autoclaved separately under 80% N₂ and 20% CO₂ gas mixture and solution C is autoclaved under 100% N₂ gas. To complete the medium appropriate amounts of solutions B and C are added to solution A. Adjust pH of the complete medium to 5.7. After inoculation pressurize vials to 2 bar overpressure with sterile 80% H₂ and 20% CO₂ gas mixture.

Note: Solution A has to be prepared freshly. It cannot be stored for a long period of time.