

**829d. DESULFUROBACTERIUM INDICUM MEDIUM**

|   |         |    |
|---|---------|----|
| Sea Salts (SIGMA)                           | 30.00   | g  |
| NH <sub>4</sub> Cl                          | 0.30    | g  |
| KH <sub>2</sub> PO <sub>4</sub>             | 0.35    | g  |
| MES [2-(N-morpholino) ethane sulfonic acid] | 1.95    | g  |
| Na-acetate                                  | 0.16    | g  |
| Trace element solution (see medium 141)     | 10.00   | ml |
| Na-resazurin solution (0.1% w/v)            | 0.50    | ml |
| Sulfur, powdered                            | 10.00   | g  |
| NaHCO <sub>3</sub>                          | 1.00    | g  |
| Vitamin solution (see medium 141)           | 10.00   | ml |
| Na <sub>2</sub> S x 9 H <sub>2</sub> O      | 0.50    | g  |
| Distilled water                             | 1000.00 | ml |

Dissolve ingredients (except sulfur, bicarbonate, vitamins and sulfide), boil medium for 1 min, then cool to room temperature under 80% H<sub>2</sub> and 20% CO<sub>2</sub> gas atmosphere. Adjust pH to 6.0 and dispense under 80% H<sub>2</sub> and 20% CO<sub>2</sub> gas atmosphere into anoxic Hungate-type tubes or serum vials which contain already the appropriate amount of sulfur. Autoclave at a temperature of **110°C** for 20 min! Add vitamins and sulfide from sterile anoxic stock solutions prepared under 100% N<sub>2</sub> gas atmosphere and bicarbonate from a sterile anoxic stock solution prepared under 80% N<sub>2</sub> and 20% CO<sub>2</sub> gas mixture. Vitamins are sterilized by filtration. Adjust pH of the complete medium to 6.0 – 6.5, if necessary. After inoculation add 1 bar overpressure of sterile 80% H<sub>2</sub> and 20% CO<sub>2</sub> gas mixture.