

1665. DESULFAMPLUS MEDIUM

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| NaCl | 20.0 | g |
| MgCl ₂ x 6 H ₂ O | 6.0 | g |
| Na ₂ SO ₄ | 3.0 | g |
| CaCl ₂ x 2 H ₂ O | 0.2 | g |
| KCl | 0.5 | g |
| NH ₄ Cl | 0.3 | g |
| Wolfe's mineral elixir (see medium 792) | 2.0 | ml |
| HEPES (SIGMA) | 2.4 | g |
| Na ₂ -fumarate | 1.0 | g |
| Yeast extract (OXOID) | 0.2 | g |
| Na-resazurin solution (0.1% w/v) | 0.5 | ml |
| 0.5 M K-phosphate buffer, pH 7.0 | 2.0 | ml |
| NaHCO ₃ | 1.0 | g |
| FeCl ₂ x 4 H ₂ O solution (0.2% w/v in 0.02 N HCl) | 10.0 | ml |
| Vitamin solution (see medium 141) | 10.0 | ml |
| L-Cysteine-HCl x H ₂ O | 0.5 | g |
| Distilled water | 1000.0 | ml |

Dissolve ingredients (except phosphate buffer, bicarbonate, ferrous chloride, vitamins and cysteine), adjust pH to 7.2 with NaOH and sparge medium with 100% N₂ gas for 30 – 45 min to make it anoxic. Dispense medium under the same gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. After autoclaving add phosphate buffer, ferrous chloride, vitamins and cysteine from sterile anoxic stock solutions prepared under 100% N₂ gas atmosphere and bicarbonate from a sterile stock solution prepared under 80% N₂ and 20% CO₂ gas mixture. Vitamins should be sterilized by filtration. Adjust pH of the complete medium to 7.2 – 7.5 , if necessary.