



Main sol. 558

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| KH ₂ PO ₄ | 1.50 | g |
| Na ₂ HPO ₄ x 12 H ₂ O | 4.20 | g |
| NH ₄ Cl | 0.50 | g |
| MgCl ₂ x 6 H ₂ O | 0.36 | g |
| Na ₂ SO ₄ | 1.00 | g |
| Modified Wolin's mineral solution | 10.00 | ml |
| Sodium resazurin (0.1% w/v) | 0.50 | ml |
| D-Glucose | 10.00 | g |
| Yeast extract | 2.00 | g |
| Wolin's vitamin solution (10x) | 1.00 | ml |
| L-Cysteine HCl x H ₂ O | 0.50 | g |
| Na ₂ S x 9 H ₂ O | 0.50 | g |
| Distilled water | 1000.00 | ml |

Dissolve ingredients (except glucose, yeast extract, vitamin solution and reducing agents), adjust pH to 3.5 using HCl and sparge medium with 80% N₂ and 20% CO₂ gas mixture for 30 - 45 min to make it anoxic. Dispense medium under same gas atmosphere into anoxic Hungate-type tubes or serum vials. After autoclaving add glucose, yeast extract, vitamin solution, cysteine and sulfide from sterile anoxic stock solutions prepared under 100% N₂ gas. The vitamin solution should be sterilized by filtration. Adjust pH of complete medium to 3.5, if necessary.