

335. EUBACTERIUM OXIDOREDUCENS MEDIUM**Solution A:**

Mineral solution (see below)	50.00	ml
Trace element solution SL-10 (see medium 320)	1.00	ml
Yeast extract	2.00	g
Crotonic acid	5.00	g
Vitamin solution (see medium 503)	1.00	ml
Resazurin	1.00	mg
Distilled water	840.00	ml

Adjust pH to 6.9 before autoclaving.

Solution B:

NaHCO ₃	5.00	g
Distilled water	100.00	ml

Solution C:

L-Cysteine-HCl x H ₂ O	0.24	g
Distilled water	10.00	ml

Solution D:

Na ₂ S x 9 H ₂ O	78.00	mg
Distilled water	1.00	ml

Prepare solution A under 80% N₂ and 20% CO₂ gas mixture, distribute 8.9 ml per Hungate-type tube under same gas atmosphere and autoclave. Filter sterilize NaHCO₃ solution and gas for 20 min with 80% N₂ and 20% CO₂ gas mixture. Autoclave cysteine and sulfide under 100% N₂ gas. Complete medium by adding the solutions B, C, and D with a syringe to solution A. Adjust final pH of medium to 7.0 - 7.5.

Mineral solution:

KH ₂ PO ₄	10.0	g
MgCl ₂ x 6 H ₂ O	6.6	g
NaCl	8.0	g
NH ₄ Cl	8.0	g
CaCl ₂ x 2 H ₂ O	1.0	g
Distilled water	1000.0	ml