



198: JCM MEDIUM No. 198

Final pH: 7.4

Final volume: 1019 ml

KH ₂ PO ₄	0.75	g
K ₂ HPO ₄	0.75	g
MnCl ₂ x 4 H ₂ O	0.36	g
NH ₄ Cl	1.00	g
NaHCO ₃	4.00	g
Trace mineral solution	9.00	ml
Trace vitamins	10.00	ml
Na ₂ S x 9 H ₂ O	0.50	g
L-Cysteine HCl x H ₂ O	0.50	g
Resazurin	1.00	mg
Distilled water	1000.00	ml

Mix ingredients, except NaHCO₃, Trace vitamins, Na₂S x 9H₂O and L-cysteine x HCl x H₂O, and autoclave under a H₂-CO₂ (80:20) atmosphere. Filter-sterilize NaHCO₃ as 8% solution and Trace vitamins while gassing with N₂. Separately autoclave Na₂S x 9H₂O and L-cysteine x HCl x H₂O as 5% solutions under a N₂ atmosphere. Aseptically and anaerobically add the NaHCO₃ solution and Trace vitamins to the medium under a H₂-CO₂ (80:20) atmosphere. Prior to inoculation, add sterile the Na₂S x 9H₂O and L-cysteine x HCl solutions, and adjust pH to 7.4. Pressurize inoculated bottles to 200 kPa H₂-CO₂ (80:20).

Trace mineral solution

Nitrilotriacetic acid	4.500	g
FeCl ₂ x 4 H ₂ O	0.400	g
MnCl ₂ x 4 H ₂ O	0.100	g
CoCl ₂ x 6 H ₂ O	0.170	g
ZnCl ₂	0.100	g
CaCl ₂ x 2 H ₂ O	0.020	g
H ₃ BO ₃	0.019	g
Na ₂ MoO ₄ x 2 H ₂ O	0.010	g
NiCl ₂ x 6 H ₂ O	0.030	g
Distilled water	1000.000	ml

Adjust pH to 7.0 with 1 N KOH.

Trace vitamins

Biotin	2.00	mg
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Folic acid	2.00	mg
Pyridoxine hydrochloride	10.00	mg
Thiamine HCl	5.00	mg
Riboflavin	5.00	mg
Nicotinic acid	5.00	mg
Calcium pantothenate	5.00	mg
Vitamin B ₁₂	0.10	mg
p-Aminobenzoic acid	5.00	mg
Lipoic acid	5.00	mg
Distilled water	1000.00	ml