

507b: CARBOXYDOCELLA MANGANICA MEDIUM

KCl	0.33	g
MgCl ₂ × 6 H ₂ O	0.52	g
CaCl ₂ × 2 H ₂ O	0.29	g
NH ₄ Cl	0.33	g
KH ₂ PO ₄	0.33	g
Trace element solution SL-4	10.00	ml
Na ₂ CO ₃	2.50	g
Na-pyruvate	1.00	g
Yeast extract	0.20	g
Wolin's vitamin solution (10x)	1.00	ml
Distilled water	1000.00	ml

Dissolve ingredients except carbonate, pyruvate, yeast extract, and vitamins, then sparge medium with 100% CO₂ gas for 30 - 45 min to make it anoxic. Add carbonate and adjust pH to 6.0 - 6.5, then dispense medium under 100% CO₂ gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. Add pyruvate, yeast extract and vitamins from sterile anoxic stock solutions prepared under 100% N₂ gas atmosphere. Stock solutions of pyruvate and vitamins are sterilized by filtration. After completing the medium adjust pH to 6.5 with a sterile anoxic stock solution of 1 N NaOH.

Trace element solution SL-4 (from medium 14)

Na ₂ -EDTA	0.50	g
FeSO ₄ × 7 H ₂ O	0.20	g
ZnSO ₄ × 7 H ₂ O	0.10	g
MnCl ₂ × 4 H ₂ O	0.03	g
H ₃ BO ₃	0.30	g
CoCl ₂ × 6 H ₂ O	0.20	g
CuCl ₂ × 2 H ₂ O	0.01	g
NiCl ₂ × 6 H ₂ O	0.02	g
Na ₂ MoO ₄ × 2 H ₂ O	0.03	g
Distilled water	1000.00	ml

First dissolve EDTA in distilled water and adjust pH to 7.0 using 2 N NaOH; then add other compounds.

Wolin's vitamin solution (10x) (from medium 120)

Biotin	20.00	mg
Folic acid	20.00	mg
Pyridoxine hydrochloride	100.00	mg
Thiamine HCl	50.00	mg

Microorganisms

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Riboflavin	50.00	mg
Nicotinic acid	50.00	mg
Calcium D-(+)-pantothenate	50.00	mg
Vitamin B ₁₂	1.00	mg
p-Aminobenzoic acid	50.00	mg
(DL)-alpha-Lipoic acid	50.00	mg
Distilled water	1000.00	ml