1525: KALLOTENUE MEDIUM

HEPES buffer (SIGMA)	3.57	g
NH_4Cl	27.00	mg
$CaSO_4$ x 2 H_2O	60.00	mg
$MgSO_4$ x 7 H_2O	0.10	g
NaCl	8.00	mg
KNO_3	0.10	g
$NaNO_3$	0.69	g
Na_2HPO_4	0.11	g
Modified Wolin's mineral solution	10.00	ml
D-Glucose	2.00	g
Trypticase peptone (BD BBL)	1.00	g
Wolin's vitamin solution (10x)	1.00	ml
Distilled water	1000.00	ml

Dissolve ingredients (except glucose, peptone and vitamins) and adjust pH to 8.1. Dispense medium under air atmosphere into culture vessels suitable for anaerobic cultivation (e.g., 10 ml medium per Balch-type tube), seal and autoclave. Add glucose, peptone and vitamins from sterile stock solutions sterilized by filtration. Adjust pH of complete medium to 7.5, if necessary.

Modified Wolin's mineral solution (from medium 141)

Nitrilotriacetic acid	1.50	g
$MgSO_4$ x 7 H_2O	3.00	g
$MnSO_4$ x H_2O	0.50	g
NaCl	1.00	g
$FeSO_4$ x 7 H_2O	0.10	g
$CoSO_4$ x 7 H_2O	0.18	g
$CaCl_2$ x 2 H_2O	0.10	g
$ZnSO_4$ x 7 H_2O	0.18	g
$CuSO_4$ x 5 H_2O	0.01	g
$AlK(SO_4)_2$ x 12 H_2O	0.02	g
H_3BO_3	0.01	g
Na_2MoO_4 x 2 H_2O	0.01	g
$NiCl_2$ x 6 H_2O	0.03	g
Na_2SeO_3 x 5 H_2O	0.30	mg
Na_2WO_4 x 2 H_2O	0.40	mg
Distilled water	1000.00	ml

First dissolve nitrilotriacetic acid and adjust pH to 6.5 with KOH, then add minerals. Adjust final to pH 7.0 with KOH.

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
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