

1633. V2 MEDIUM

Peptone (Bacto)	0.250	g
Yeast extract (Bacto)	0.250	g
HEPES	2.380	g
Mineral salt solution (see below)	20.000	ml
Trace element solution (see below)	1.000	ml
NaHCO ₃	0.025	g
CaCO ₃	0.025	g
CaCl ₂ x 2 H ₂ O	0.015	g
MgSO ₄ x 7 H ₂ O	0.049	g
KNO ₃	0.010	g
KH ₂ PO ₄	1.400	mg
NH ₄ Cl	0.530	mg
Agar (Bacto)	15.00	g
Double distilled water	730.00	ml

Sterilize 20 min./121°C. After cooling to 60°C add:

N-Acetylglucosamine (10% stock solution)	8.0	ml
Glucose sol. (2.5%, sterile-filtered)	10.0	ml
Vitamin sol. (double conc.)(see below, medium 621)	5.0	ml

Adjust pH to 7.0 with 40% KOH.

Mineral salts solution:

Nitrilotriacetic acid (NTA)	10.00	g
MgSO ₄ x 7 H ₂ O	29.70	g
CaCl ₂ x 2 H ₂ O	3.34	g
Na ₂ MoO ₇ O ₄ x 2 H ₂ O	12.67	mg
FeSO ₄ x 7 H ₂ O	99.00	mg
"Metals 44" (see below)	50.00	ml
Distilled water	950.00	ml

Dissolve the nitrilotriacetic acid, adjust the pH to 7.0 with KOH (about 7.3 g). Dissolve other salts separately, combine and adjust the pH to 6.8 with NaOH or H₂SO₄.

"Metals 44":

Na-EDTA	250.000	mg
ZnSO ₄ x 7 H ₂ O	1095.000	mg
FeSO ₄ x 7 H ₂ O	500.000	mg
MnSO ₄ x H ₂ O	154.000	mg
CuSO ₄ x 5 H ₂ O	39.200	mg
Co(NO ₃) ₂ x 6 H ₂ O	24.800	mg
Na ₂ B ₄ O ₇ x 10 H ₂ O	17.700	mg
Distilled water	1000.000	ml

Trace element solution:

Na-Nitrilotriacetat	1.50	g
MnSO ₄ x H ₂ O	0.50	g
Fe SO ₄ x 7 H ₂ O	0.10	g
Co(NO ₃) ₂ x 6 H ₂ O	0.10	g
ZnCl ₂	0.10	g
NiCl ₂ x 6 H ₂ O	0.05	g
H ₂ SeO ₃	0.05	g
CuSO ₄ x 5 H ₂ O	0.01	g
AlK(SO ₄) ₂ x 12 H ₂ O	0.01	g
H ₃ BO ₃	0.01	g
Na ₂ MoO ₄ x 2 H ₂ O	0.01	g
Na ₂ WO ₄ x 2 H ₂ O	0.01	g
Double distilled water	1000.00	ml

Vitamin solution (double conc.):

Biotin	4.00	mg
Folic acid	4.00	mg
Pyridoxine-HCl	20.00	mg
Riboflavine	10.00	mg
Thiamine-HCl x 2 H ₂ O	10.00	mg
Nicotinamide	10.00	mg
D-Ca-pantothenate	10.00	mg
Vitamin B ₁₂	0.20	mg
p-Aminobenzoic acid	10.00	mg
Double distilled water	1000.00	ml