

ATCC medium: 509 *Prosthecomicrobium* and *Ancalomicromicrobium* medium with nicotinamide

Prepare Medium 289 (see below) as directed with the addition of 5.0 mg of nicotinamide to the Vitamin Solution.

ATCC Medium 289:

Ammonium sulfate.....	0.25 g
Glucose.....	0.25 g
Vitamin Solution (see below)	10.0 ml
Modified Hutner's Basal Salts (see below).....	20.0 ml
Na ₂ HPO ₄	0.071 g
Distilled water to.....	1.0 L

Vitamin Solution:

Biotin.....	2.0 mg
Folic acid.....	2.0 mg
Thiamine . HCl.....	5.0 mg
Calcium D-(+)-pantothenate....	5.0 mg
Vitamin B12.....	0.1 mg
Riboflavin.....	5.0 mg
Distilled water.....	1.0 L

Modified Hutner's Basal Salts:

Nitritotriacetic acid.....	10.0 g
MgSO ₄ . 7H ₂ O	29.7 g
CaCl ₂ . 2H ₂ O	3.34 g
Ammonium molybdate.....	9.25 mg
FeSO ₄ . 7H ₂ O	99.0 mg
Metals "44" (see below)	50.0 ml
Distilled water to.....	1.0 L

Dissolve and neutralize the nitritotriacetic acid with KOH (7.3 g); add the other ingredients and adjust the pH to 6.6 - 6.8 before bringing the volume to 1.0 L with distilled water.

Metals "44":

EDTA.....	0.25 g
ZnSO ₄ . 7H ₂ O.....	1.1 g
FeSO ₄ . 7H ₂ O.....	0.5 g
MnSO ₄ . 7H ₂ O.....	0.154 g
CuSO ₄ . 5H ₂ O.....	0.04 g
Co(NO ₃) ₂ . 6H ₂ O.....	0.025 g
Na ₂ B ₄ O ₇ . 10H ₂ O.....	0.018 g
Distilled water.....	100.0 ml

Initially add a few drops of H₂SO₄ to the distilled water to retard precipitation.