

ATCC Medium: 2815 Heliorestis Medium

KH ₂ PO ₄	0.5 g
NaCl.....	0.5 g
NgSO ₄ ·7H ₂ O.....	0.2 g
CaCl ₂ ·2H ₂ O.....	0.075 g
Na-acetate.....	1.0g
Na-pyruvate.....	1.0 g
Yeast Extract.....	0.10 g
Trace Elements Solution SL-6 (<i>see below</i>).....	1.0 ml
Vitamin B ₁₂	20 ug
Biotin.....	20 ug
BICINE buffer (Sigma).....	final concentration of 10mM
DI Water.....	1000.0 ml

Dissolve ingredients in a total of 930 ml distilled water and sterilize by autoclaving. After autoclaving add sterile solutions of NaHCO₃, Na₂S·9H₂O, and NH₄Cl (see below). Adjust the pH 9.0 – 9.2 with the addition of sterile sodium hydroxide. Distribute medium into sterile screw cap test tube, fill to capacity.

NH₄CL Solution

0.5g dissolved in 10 ml distilled water and filter sterilize.

NaHCO₃ Solution

Place 3 g in a flask and autoclave dry, then dissolve in 50 ml sterile distilled water.

Na₂S·9H₂O Solution

0.5g dissolved in 10 ml distilled water and then filter sterilized

Trace Elements Solution SL-6 (ATCC MD-TMS can be substituted)

ZnSO ₄ ·7H ₂ O.....	0.10 g
MnCl ₂ ·4H ₂ O.....	0.03 g
H ₃ BO ₃	0.3 g
CoCl ₂ ·6H ₂ O.....	0.2 g
CuCl ₂ ·2H ₂ O.....	0.01 g
NiCl ₂ ·6H ₂ O.....	0.02 g
Na ₂ MoO ₄ ·H ₂ O.....	0.03 g
DI Water.....	1000.0 ml

Adjust final pH of Trace Elements Solution SL-6 to 3.4.