

ATCC medium: 1482 PYG plus 0.1% Tween 80

Tween 80.....	1.0 g
Glucose.....	10.0 g
Peptone (BD 211677).....	5.0 g
Trypticase Peptone (BD 211921).....	5.0 g
Yeast extract.....	10.0 g
0.025% Resazurin.....	4.0 ml
Hemin Solution (see below).....	10.0 ml
Salts Solution (see below).....	40.0 ml
Vitamin K1 Solution (see below).....	0.2 ml
L-Cysteine . HCl.....	0.5 g
Distilled water.....	950.0 ml

Bring medium without cysteine to a boil; add cysteine; adjust medium for a final pH of 7.0; autoclave at 121C for 15 minutes.

Hemin Solution:

Hemin.....	50.0 mg
N NaOH.....	1.0 ml
Distilled water to.....	100.0 ml

Dissolve hemin in NaOH. Add distilled water. Autoclave at 121C for 15 minutes.

Salts Solution:

CaCl ₂	0.2 g
MgSO ₄	0.2 g
K ₂ HPO ₄	1.0 g
KH ₂ PO ₄	1.0 g
NaHCO ₃	10.0 g
NaCl.....	2.0 g
Distilled water.....	1.0 L

Vitamin K1 Solution:

Vitamin K1.....	0.15 ml
95% Ethanol.....	30.0 ml

Store K1 solution in a brown bottle under refrigeration. Discard solution after 1 month.