

ATCC medium: 1539 Modified chopped meat medium (ATCC medium 1490) with formate and fumarate

Prepare a solution containing sodium formate and fumaric acid at a concentration of 6% each, adjust the pH to 7.0 and filter-sterilize. Prior to inoculation add 0.25 ml to a tube containing approximately 5 ml of Medium 1490 (see below).

ATCC Medium 1490:

Ground beef (fat-free).....500.0 g
Distilled water.....1.0 L
N NaOH.....25.0 ml

Mix meat, water and NaOH and bring to a boil while stirring. Cool to room temperature, skim fat from surface, and filter, retaining both meat particles and filtrate. Add sufficient distilled water to filtrate to restore volume to 1.0 L.

To this filtrate, add:

Trypticase Peptone (BD 211921)....30.0 g
Yeast extract.....5.0 g
K₂HPO₄5.0 g
0.025% Resazurin.....4.0 ml
Agar (if necessary).....20.0 g

Boil and cool medium under 80% N₂, 10% H₂, 10% CO₂, and add:

L-Cysteine . HCl.....0.5 g
Hemin Solution (see below).....10.0 ml
Vitamin K1 Solution (see below)....0.2 ml

Adjust medium for final pH 7.0. Anaerobically dispense 7 ml into tubes containing meat particles (1 part meat to 5 parts fluid) under the same gas phase.

Vitamin K1 Solution:

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

Store solution in brown bottle under refrigeration. Discard after one month.

Hemin Solution:

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

Autoclave at 121C for 15 minutes.