

ATCC medium: 1943 *Fibrobacter* medium

KH ₂ PO ₄	0.3 g
K ₂ HPO ₄	0.3 g
(NH ₄) ₂ SO ₄	0.3 g
NaCl.....	0.6 g
MgSO ₄ . 7H ₂ O	0.12 g
CaCl ₂ . 2H ₂ O	0.08 g
Tryptone (BD 211705).....	1.0 g
Yeast extract.....	0.5 g
Resazurin.....	1.0 mg
Cellobiose.....	4.0 g
Vitamin Solution (see below)	20.0 ml
Trace Elements (see below).....	1.0 ml
VFA Solution (see below)	4.65 ml
Na ₂ CO ₃	4.0 g
Distilled water.....	960.0 ml

Heat medium without sodium carbonate to boiling and cool under a stream of 100% CO₂. Add sodium carbonate and equilibrate medium under same gas phase for approximately 10 minutes. Dispense anaerobically into desired containers. Autoclave at 121C for 15 minutes.

Prior to inoculation, add the following reducing agents which have been prepared as stock solutions under nitrogen and autoclaved:

2.5% L-Cysteine . HCl.....	10.0 ml
2.5% Na ₂ S . 9H ₂ O	10.0 ml

Final pH of completed medium is 6.6 +/- 0.1.

Vitamin Solution:

Lipoic acid.....	20.0 mg
Thiamine . HCl.....	20.0 mg
Calcium D-(+)-pantothenate..	20.0 mg
Nicotinamide.....	20.0 mg
Riboflavin.....	20.0 mg
Pyridoxal hydrochloride.....	20.0 mg
Pyridoxamine . 2HCl.....	20.0 mg
p-Aminobenzoic acid.....	1.0 mg
Biotin.....	1.0 mg
Cyanocobalamin.....	1.0 mg
Distilled water.....	100.0 ml

Trace Elements:

ZnSO ₄ . 7H ₂ O	0.1 g
MnCl ₂ . 4H ₂ O	0.03 g
H ₃ BO ₃	0.2 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 . 2H ₂ O.....	0.03 g
FeSO ₄ . 7H ₂ O	2.0 g
Distilled water.....	1.0 L

VFA Solution:

Acetic acid.....	17.0 ml
Propionic acid.....	6.0 ml
n-Butyric acid.....	4.0 ml
n-Valeric acid.....	1.0 ml
Isovaleric acid.....	1.0 ml
Isobutyric acid.....	1.0 ml
DL-alpha-Methylbutyric acid..	1.0 ml
Distilled water to.....	310.0 ml

Add volatile fatty acids to approximately 200 ml distilled water.
Neutralize to pH 7.0 with NaOH pellets. Bring volume to 310 ml.