

Main sol. J1312

1. Mix ingredients, bring to a boil and cool down under a N_2 - CO_2 (4:1, v/v) gas stream and autoclave under the same gas mixture. After cooling, aseptically and anaerobically add per liter the following solutions (filter-sterilized):

NH_4Cl	1.000	g
$NaCl$	2.100	g
$MgCl_2 \times 6 H_2O$	0.825	g
$CaCl_2 \times 2 H_2O$	0.100	g
KH_2PO_4	0.500	g
Trace element solution SL-10	1.000	ml
Selenite-tungstate solution	1.000	ml
Yeast extract (BD-Difco)	1.000	g
Resazurin (0.1%)	0.500	ml
Trace vitamins	10.000	ml
$NaHCO_3$ (8%)	30.000	ml
Taurine (0.5 M)	20.000	ml
Sodium pyruvate (1 M)	20.000	ml
Sodium L-lactate (0.5 M)	20.000	ml
Hemin solution	10.000	ml

2. Prior to inoculation, aseptically and anaerobically add 0.01 volume of L-cysteine x HCl x H_2O to the medium (3% solution, autoclaved and stored under a N_2 atmosphere).