Microorganisms



395b: FERVIDICOCCUS MEDIUM

NH₄Cl	0.33	g
KH ₂ PO ₄	0.33	g
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KCI	0.33	g
CaCl ₂ x 2 H ₂ O	0.44	g
$MgCl_2 \times 6 H_2O$	0.70	g
NaCl	0.50	g
Trace element solution SL-10	1.00	ml
Yeast extract (OXOID)	0.50	g
Sodium resazurin (0.1% w/v)	0.50	ml
NaHCO ₃	0.80	g
Trypticase peptone (BD BBL)	2.00	g
Wolin's vitamin solution (10x)	1.00	ml
$Na_2S \times 9 H_2O$	0.50	g
Distilled water	1000.00	ml

Dissolve ingredients (except bicarbonate, Trypticase, vitamins and sulfide), then sparge medium with $80\%~N_2$ and $20\%~CO_2$ gas mixture for 30 - 45 min to make it anoxic. Dispense medium under same gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. After autoclaving add Trypticase, vitamins and sulfide from sterile anoxic stock solutions prepared under $100\%~N_2$ gas atmosphere and bicarbonate from a sterile anoxic stock solution prepared under $80\%~N_2$ and $20\%~CO_2$ gas mixture. Vitamins are sterilized by filtration. Adjust pH of complete medium to 6.0 - 6.1, if necessary.

For <u>DSM 16532</u>: Replace Trypticase peptone with 5.00 g/l soluble starch as substrate.

Trace element solution SL-10 (from medium 320)

HCl (25%)	10.00	ml
FeCl ₂ x 4 H ₂ O	1.50	g
ZnCl ₂	70.00	mg
$MnCl_2 \times 4 H_2O$	100.00	mg
H_3BO_3	6.00	mg
CoCl ₂ x 6 H ₂ O	190.00	mg
CuCl ₂ x 2 H ₂ O	2.00	mg
$NiCl_2 \times 6 H_2O$	24.00	mg
$Na_2MoO_4 \times 2 H_2O$	36.00	mg
Distilled water	990.00	ml

First dissolve $FeCl_2$ in the HCl, then dilute in water, add and dissolve the other salts. Finally make up to 1000.00 ml.

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Wolin's vitamin solution (10x) (from medium 120)

Biotin	20.00	mg
Folic acid	20.00	mg
Pyridoxine hydrochloride	100.00	mg
Thiamine HCl	50.00	mg
Riboflavin	50.00	mg
Nicotinic acid	50.00	mg
Calcium D-(+)-pantothenate	50.00	mg
Vitamin B ₁₂	1.00	mg
p-Aminobenzoic acid	50.00	mg
(DL)-alpha-Lipoic acid	50.00	mg
Distilled water	1000.00	ml