171. THERMOANAEROBACTER KIVUI MEDIUM

K_2HPO_4	0.22	g
KH_2PO_4	0.22	g
NaH_2PO_4 x H_2O	4.50	g
Na_2HPO_4 x 12 H_2O	6.10	g
NH_4Cl	0.31	g
$(NH_4)_2SO_4$	0.22	g
NaCl	0.45	g
$MgSO_4$ x 7 H_2O	0.09	g
$CaCl_2$ x 2 H_2O solution (0.1% w/v)	6.00	ml
$FeSO_4$ x 7 H_2O solution (0.1% w/v in 0.1 N H_2SO_4)	2.00	ml
Trace element solution (see medium 141)	10.00	ml
Na-resazurin solution (0.1% w/v)	0.50	ml
L-Cysteine-HCl x H_2O	0.50	g
Na_2S x 9 H_2O	0.50	g
Distilled water	1000.00	ml

Dissolve ingredients except cysteine and sulfide, sparge medium with 80% H_2 and 20% CO_2 gas mixture for 30 – 45 min to make it anoxic, then distribute under same gas atmosphere into Hungate-type tubes or serum vials and autoclave. Add cysteine and sulfide from sterile anoxic stock solutions prepared under 100% N_2 gas atmosphere. Adjust pH of complete medium to 6.5. After inoculation pressurize cultivation vessels to one atmosphere overpressure with sterile 80% H_2 and 20% CO_2 gas mixture.