88c: ZESTOSPHAERA MEDIUM

Final pH: 6.0
Final volume: 1010 ml

$(NH_4)_2SO_4$	1.30	g
KH_2PO_4	0.28	g
$MgSO_4$ x 7 H_2O	0.25	g
$CaCl_2$ x 2 H_2O	0.07	g
$FeCl_3$ x 6 H_2O	0.02	g
Allen's trace element solution	10.00	ml
$Na_2S_2O_3$ x 5 H_2O	2.00	g
Yeast extract (OXOID)	1.00	g
Tryptone peptone (Bacto BD)	1.00	g
Casamino acids (Difco BD)	0.50	g
Na_2S x 9 H_2O	0.50	g
Distilled water	1000.00	ml

Dissolve ingredients (except thiosulfate, yeast extract, Tryptone, Casamino acids and sulfide), sparge medium with 80% N_2 and 20% CO_2 gas mixture to make it anoxic and dispense medium under same gas atmosphere into serum bottels. After autoclaving add thiosulfate, yeast extract, Tryptone, Casamino acids and sulfide from sterile anoxic stock solutions prepared under 100% N_2 gas. Prior to inoculation check pH and adjust to 6.0, if necessary.

Allen's trace element solution (from medium 88)

$MnCl_2$ x 4 H_2O	180.00	mg
$Na_2B_4O_7$ x 10 H_2O	450.00	mg
$ZnSO_4$ x 7 H_2O	22.00	mg
$CuCl_2$ x 2 H_2O	5.00	mg
Na_2MoO_4 x 2 H_2O	3.00	mg
$VOSO_4$ x 2 H_2O	3.00	mg
$CoSO_4$ x 7 H_2O	1.00	mg
Distilled water	1000.00	ml

Adjust pH of final solution to 2 with 1 N HCl.