

1210. GEOGLOBUS MEDIUM

NaCl	18.00	g
MgCl ₂ x 6 H ₂ O	4.00	g
KCl	0.33	g
CaCl ₂ x 2 H ₂ O	0.33	g
(NH ₄) ₂ SO ₄	0.50	g
Trace element solution SL-10 (see medium 320)	1.00	ml
Selenite-tungstate solution (see medium 385)	1.00	ml
NaHCO ₃	2.50	g
Na-acetate	1.50	g
KH ₂ PO ₄	0.33	g
Yeast extract	0.20	g
Vitamin solution (see medium 141)	1.00	ml
Amorphous Fe(OH) ₃ sludge (see below)	200.00	ml
Distilled water	800.00	ml

Dissolve ingredients except bicarbonate, acetate, hydrogenphosphate, yeast extract, vitamins and ferric iron hydroxide sludge. Suspend pellet of ferric iron hydroxide in medium and sparge with 80% N₂ and 20% CO₂ gas mixture for 30 – 45 min to make it anoxic. Thereafter, dispense suspension under 80% N₂ and 20% CO₂ gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave for 30 min. Add acetate, hydrogenphosphate, yeast extract and vitamins (sterilized by filtration) from sterile anoxic stock solutions prepared under 100% N₂ gas and bicarbonate from a sterile anoxic stock solution prepared under 80% N₂ and 20% CO₂ gas mixture. The pH of the complete medium should be at 6.5 – 6.8.

Amorphous Fe(OH)₃:

Slowly titrate 320 ml of a FeCl₃ x 6 H₂O stock solution (60.00 g/l) with 10% (w/v) NaOH to pH 8.0-8.5 under agitation (use magnetic stirrer). Total amount of added NaOH approx. 80 – 100 ml. The precipitated Fe(OH)₃ should be stored at room temperature overnight with surface covered with water. Thereafter, centrifuge at 2000 rpm for 5 min and discard the supernatant. Resuspend the pellet in medium as described above.