Main sol. 916

Solution A	912.00	ml
Solution B	50.00	ml
Solution C	25.00	ml
Solution D	10.00	ml
Solution E	1.00	ml
Solution F	10.00	ml

1. Sparge solution A with 80% N_2 and 20% CO_2 gas mixture for 30 – 45 min to make it anoxic, then dispense under same gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. Solutions B and D are autoclaved under 100% N_2 gas atmosphere. Solution C is autoclaved separately under 80% N_2 and 20% CO_2 gas atmosphere. Solutions E and F are prepared under 100% N_2 gas and sterilized by filtration. To complete the medium appropriate amounts of solutions B to F are added to the sterile solution A in the sequence as indicated. The pH of the complete medium should be 7.0 - 7.2.

2. Note: Addition of 10 - 20 mg sodium dithionite per liter (e.g. from 5% (w/v) solution, freshly prepared under N_2 and filter-sterilized) may stimulate growth at the beginning.