

Main sol. 717

$(\text{NH}_4)\text{H}_2\text{PO}_4$	2.88	g
$\text{MgSO}_4 \times 7 \text{ H}_2\text{O}$	0.10	g
$\text{Ca}(\text{NO}_3)_2 \times 4 \text{ H}_2\text{O}$	0.05	g
Modified Wolin's mineral solution	10.00	ml
Yeast extract	0.10	g
Sodium resazurin (0.1% w/v)	0.50	ml
Na_2CO_3	1.50	g
Na-L-lactate	5.00	g
Na_2 -fumarate	3.20	g
Wolin's vitamin solution (10x)	1.00	ml
Seven vitamins solution	1.00	ml
$\text{Na}_2\text{S} \times 9 \text{ H}_2\text{O}$	0.10	g
Distilled water	1000.00	ml

Dissolve ingredients except carbonate, lactate, fumarate, vitamins and sulfide. Sparge medium 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. Add lactate, fumarate, vitamins and sulfide from sterile anoxic stock solutions prepared under 100% N_2 gas and carbonate from a sterile anoxic stock solution prepared under 80% N_2 and 20% CO_2 gas atmosphere. Stock solutions of fumarate and vitamins should be sterilized by filtration. The pH of the complete medium should be at 7.0 - 7.2.