



Supplementary Material

Utilization of Emulsiflex Prepared *Escherichia coli* Strain EC4 Oxyrase for Improved Cultivation of Anaerobic Bacteria by using Hungate Technique and Determination of its Kinetic Parameters

Muhammad Usman Ahmad and Ikram-ul-Haq*

Institute of Industrial Biotechnology, GC University, Lahore-54000

* Corresponding author: ikmhaq@yahoo.com

0030-9923/2019/0003-0825 \$ 9.00/0

Copyright 2019 Zoological Society of Pakistan

Supplementary Table I.- Medium composition for *Anaerobaculum hydrogeniformans* OS1 (1.0 L).

Components	Quantity
[†] Wolin's Trace Elements	10mL
*OS1 Minerals Solution	10mL
[‡] Vitamins Solution	10mL
NaCl	10.0g
Yeast Extract	2.0g
TES {2-(2-[hydroxy-1,1-bis(hydroxymethyl)ethyl]amino) ethane sulfonic acid} Buffer Powder	10.0g
[†]Wolin's trace elements for 1000mL (filter sterilized)	
Ethylenediaminetetraacetic acid (EDTA)	0.50g
Magnesium sulfate heptahydrate (MgSO ₄ ·7H ₂ O)	3.0g
Manganese sulfate hydrate (MnSO ₄ ·H ₂ O)	0.50g
Sodium chloride (NaCl)	1.0g
Calcium chloride dihydrate (CaCl ₂ ·2H ₂ O)	0.10g
Cobalt(II) Chloride Hexahydrate (CoCl ₂ ·6H ₂ O)	0.10g
Zinc Sulfate Heptahydrate (ZnSO ₄ ·7H ₂ O)	0.10g
Ferrous sulfate heptahydrate (FeSO ₄ ·7H ₂ O)	0.10g
Copper sulfate pentahydrate (CuSO ₄ ·5H ₂ O)	0.01g
Sodium Molybdate Dihydrate (Na ₂ MoO ₄ ·2H ₂ O)	0.01g
Sodium selenate (Na ₂ SeO ₄)	0.01g
Sodium tungstate dihydrate (Na ₂ WO ₄ ·2H ₂ O)	0.01g
*OS1 minerals solution for 1000mL	
Sodium Chloride (NaCl)	80.0g
Ammonium Chloride (NH ₄ Cl)	100g
Potassium Chloride (KCl)	10.0g
Monopotassium phosphate (KH ₂ PO ₄)	10.0g
Magnesium sulfate heptahydrate (MgSO ₄ ·7H ₂ O)	20.0g
Calcium chloride dihydrate (CaCl ₂ ·2H ₂ O)	4.0g
[‡]Vitamins solution for 1000mL (Filter Sterilized)	
Biotin	2.0mg
Folic Acid	2.0mg
Pyridoxine-HCl	10.0mg

Components	Quantity
Thiamine-HCl	5.0mg
Riboflavin	5.0mg
Nicotinic acid	5.0mg
DL calcium Panthothenate	5.0mg
Vitamin B12	0.10mg
Para-aminobenzoic acid	5.0mg
Lipoic acid	5.0mg
Mercaptoethane sulfonic acid	5.0mg

Supplementary Table II.- Medium composition for *Akkermansia muciniphila* (1.0 L).

Components	Quantity
BBL Columbia broth powder	35gm
Hog Gastric Mucin Type III (0.25%)	0.5gm

Supplementary Table III.- Medium composition for *Biophila wadsworthia* (1.0 L).

Components	Quantity
Brucella Broth	28.0g
1% Taurine (2-aminoethanesulfonic acid)	10.0g
1mg/L Vitamin K1 (50mg/mL stock solution)	0.1mL
Resazurin	20 μ L

Supplementary Table IV.- Medium composition for *Roseburia intestinalis* (1.0 L).

Components	Quantity
Glucose	5.0g
Sodium Acetate Trihydrate ($\text{NaCH}_3\text{COO}\cdot 3\text{H}_2\text{O}$)	2.50g
Castone	10.0g
Yeast extract	2.50g
Sodium bicarbonate (NaHCO_3)	4.0g
Cysteine-HCl	1.0g
Dipotassium hydrogenphosphate (K_2HPO_4)	0.45g
Monopotassium phosphate (KH_2PO_4)	0.45g
Sodium chloride (NaCl)	0.90g
Magnesium sulfate heptahydrate ($\text{MgSO}_4\cdot 7\text{H}_2\text{O}$)	0.09g
Calcium chloride dihydrate ($\text{CaCl}_2\cdot 2\text{H}_2\text{O}$)	0.09g
Resazurin	20.0 μ L
Vitamins solutions	1.0mL
Trace elements	10.0mL
Hemin	1.0mL
Vitamin K2	10.0 μ L