APPENDIX-I

REAGENTS OF IDENTIFICATION OF ORGANISMS

1. Crystal Violet (Qualigens)

A Crystal Violet 2g
95% Alcohol 20ml

B Ammonium oxalate 0.8g
Distilled water 80ml

Mix A and B. Let it stand for 24hrs. Filter and use. If the solution is too concentrated, dilute A as much as 10 times.

2. Aqueous solution of Safranin

Safranin 0.5g
Distilled water 100ml
Store in a dark bottle

3. Lugol's Iodine

Iodine 1g
Potassium Iodide 2g
Distilled Water 300ml

The solution was stored in an amber cololured bottle.
4. Kovac's Reagent (For indole test)
   Isoamyl alcohol 150ml
   Concentrated HCl 50ml
   p-dimethyl-aminobenzaldehyde 10g

The aldehyde was dissolved in alcohol and acid was then added. It was stored in a dark coloured bottle in the refrigerator.

5. Methyl Red Reagent (for detection of acid)
   Methyl red 0.1g
   95% ethyl alcohol 300ml

The dye was dissolved in alcohol and volume made to 500ml with distilled water. Positive test is red-orange and negative is yellow.

6. a-Naphthol
   5% a-Naphthol in 95% ethyl alcohol

7. Barritt's Reagent (For Voges-Proskauer Test)
   Solution A
   a-Naphthol 6 g
   95% ethyl alcohol 100ml

   Solution B
   KOH 16 g
   Distilled water 100ml
   Store in a refrigerator

8. Benedict's Reagent (For Gluconate Test)
   Sodium Citrate 17.3 g
   Na$_2$CO$_3$ anhy. 10.0 g
   CuSO$_4$ 1.73 g
   Distilled water 100 ml
Sodium citrate and sodium carbonate were dissolved in 60ml of distilled water. Copper sulfate was dissolved in 20 ml of distilled water and added to the first solution by constant stirring. Volume was then adjusted to 100ml.

9. Basic Fuchsin

Basic fuchsin 1.0 g
Distilled water 100 ml

1 g of basic fuchsin was dissolved in distilled water and volume was made up to 100ml.

10. Nitrate Test Reagent

Dissolve 0.7g of DPA in 60ml of concentrated sulfuric acid and add 28.8 ml of distilled water. Allow to cool and add 11.3 ml of concentrated hydrochloric acid.

11. Methylene blue

Methylene blue 1 g
Distilled water 100 ml

12. Anrade's Indicator (for sugar fermentation tests)

Acid fuchsin 5 g
Distilled water 1000ml
Sodium hydroxide 150-180ml

Acid fuchsin was dissolved in distilled water and 150ml of alkali solution was added to it. The mixture was kept at room temperature with frequent shaking. More alkali was added to make the solution straw yellow.