APPENDIX

A. PREPARATION OF REAGENTS FOR PLANT ANALYSIS

(1) Reagents for Carbohydrate estimation

i) 1.5N-H$_2$SO$_4$ 20.4 ml conc. H$_2$SO$_4$ was dissolved in distilled water and volume was made up to 500 ml.

(2) Reagents for Protein estimation

i) Reagent A: 2 percent sodium carbonate + 0.1N sodium hydroxide (1:1).

ii) Reagent B: 0.5% copper sulphate + 1 percent sodium tartarate (1:1).

iii) Reagent C: Alkaline copper sulphate solution obtained by mixing 50 ml of reagent A with 1 ml of reagent B.

iv) Reagent D: Carbonate copper sulphate solution same as reagent C, except for omission of NaOH.

v) Reagent E: 1N acid Folin reagent, 100gm of sodium tungstate and 25 gm of sodium molybdate was dissolved in 700 ml of water and kept in a 1500 ml flask. 50 ml of 85% phosphoric acid and 100 ml of concentrated hydrochloric acid was added. The flask then connected with a reflux condenser and boiled gently on a heating mantle for 10 hr. At the end of the boiling period, 150 gm lithium sulphate, 50 ml of water and 3-4 drops of liquid bromine added to the flask. The reflux removed and the solution was boiled for 15 min to remove excess bromine. Cooled and diluted to 1000 ml with distilled water. The strength of this acidic solution estimated by titrating it with 1N solution of NaOH using phenolphthaleine as indicator and diluted to the required strength (1N).

vi) 1N NaOH: 4 gm of NaOH was dissolved in distilled water and finally volume was made up to 100 ml.

B. PREPARATION OF REAGENTS FOR SOIL CHEMICAL ANALYSIS

(1) For the estimation of Organic Carbon

i) 1N Potassium dichromate: 49.04 gm K$_2$Cr$_2$O$_7$ was dissolved in distilled water and finally it was made up to 1 L.
ii) 0.5N Ferrous ammonium sulphate: 0.96 g of hydrated ferrous ammonium sulphate dissolved in distilled water. 20 ml of concentrated sulphuric acid was added and volume made up to 1 L.

iii) Diphenyl amine indicator: 0.5 g diphenyl amine dissolved in a mixture of 20 ml of water and 100 ml of concentrated sulphuric acid.

iv) Orthophosphoric acid 85%

v) Sulphuric acid - Not less than 96%

(2) Nitrate nitrogen

i) Phenol disulphonic acid: 25 g of pure phenol ($C_8$H$_c$O$_h$, crystal white) taken in a dry conical flask (500 ml). 150 ml concentrated sulphuric acid (nitrate free) added and kept on boiling water bath for 2 hr. After cooling stored in amber coloured bottle.

ii) Liquor ammonia (1:1): Ammonia having 0.88 sp.gr. diluted with equal volume of water.

(3) Available phosphorus

i) Olsen's reagent: 42.0 g of NaHCO$_3$ dissolved in distilled water to give one titre of the solution and pH adjusted to 8.5 with small quantities of NaOH.

ii) Dickman and Bray's reagent: 15 g of ammonia molybdate dissolved in 300 ml of luke warm water (about 60°C), cooled and filtered. 400 ml of 10N HCl added and finally the volume made up to 1 L.

iii) Stannous chloride solution: 10 g of crystalline stannous chloride dissolved in 25 ml of concentrated HCl by warming and stored in an amber coloured bottle, giving 40% SnCl$_2$ stock solution. Just before use, 0.5 ml diluted to 66 ml with distilled water.

iv) 7N H$_2$SO$_4$: 19.6 ml concentrated sulphuric acid added to double distilled water and the final volume was made up to 100 ml.
(4) Potassium

1) Ammonium acetate solution (Neutral and Normal):
Solution of 2 N acetic acid (glacial) and 2N
ammonium hydroxide prepared by titration with
standard alkali and acid respectively and equal
volumes of the two were mixed in a large beaker.
On cooling, pH adjusted to 7.0 with acetic acid.

(5) Calcium

i) 0.01N EDTA solution: 2.0 g of ethylene diamine
tetra acetic acid dissolved in distilled water
and final volume made upto 1000 ml

ii) Murexide indicator. 0.2 g ammonium purpurate
mixed with 40 g of powdered potassium sulphate

(6) Cation exchange capacity (CEC)

1) 0.1N Sodium hydroxide solution 4 gm of sodium
hydroxide dissolved in distilled water and the
final volume was made upto 1000 ml

(7) Sulphate

1) Conditioning reagent 50 ml glycerol mixed
in a solution containing 30 ml HCl and 300
ml distilled water + 100 ml of 95% ethyl
alcohol and 75 g sodium chloride.

(8) Chloride

1) Potassium chromate indicator 50 g of potassium
chromate was dissolved in distilled water
Silver nitrate solution was added until a red
precipitate appeared . After standing
overnight filtered and diluted to 1000 ml with
distilled water

i) Standard silver nitrate titrant (0 0141N).
2.395 g silver nitrate dissolved in distilled
water and diluted to 1000 ml.

(9) Carbonates and bicarbonates

1) Phenolphthalein indicator 0 25% solution made
in 60% ethyl alcohol.
11) 0.01N Sulphuric acid 0.272 ml sulphuric acid
diluted in distilled water and final volume
made up to 100 ml

111) Methyl red indicator 0.5% solution made in
95% alcohol

(10) Nitrate nitrogen

1) Phenol disulphonic acid 25 g pure phenol (C₆H₅OH,
crystal white) in conical flask (500 ml) 150 ml
concentrated sulphuric acid (nitrate-free) and
75 ml fuming sulphuric acid (nitrate-free) added
and kept on boiling water bath for 2 hr
covered with watch glass and stored in amber
colour bottle

C. PREPARATION OF REAGENTS FOR ANALYSIS OF WATER

(1) Biological oxygen demand (BOD)

1) Manganous sulphates solution 40 gm of manganese
sulphate dissolved in distilled water and
volume was made up to 100 mL

11) Alkali azide reagent 50 g of sodium hydroxide
and 13.5 g of sodium iodide diluted to 100
ml with distilled water 1 gm of sodium azide
dissolved in 4 ml of distilled water and added
to above solution

(2) Chemical oxygen demand (COD)

1) Standard potassium dichromate solution 0.25N
12.259 g potassium dichromate dissolved in
distilled water and final volume made up to
1000 mL

11) Standard ferrous ammonium sulphate solution 0.1
N 39 g FAS dissolved in distilled water 20
ml of concentrated sulphuric acid added and
finally volume made up to 1000 mL

111) Ferroin indicator solution 1.485 g 1,10
phenanthroline monohydrate, together with 495
mg FeSO₄, dissolved in distilled water and
finally volume made up to 100 mL
3) Total hardness

i) 0.01 M ethylene diamine tetra acetic acid. 3.723 g EDTA dissolved in distilled water and diluted to 1000 mL.

ii) Eriochrome Black T indicator: 0.5 g dye was mixed with 100 g of 2,2,2 nitrilotriethanol

(4) Carbonate and bicarbonates.

i) Phenolphthalein indicator: 0.25% solution made in 60% ethyl alcohol.

ii) Standard sulphuric acid (0.01 N): 2.72 mL sulphuric acid diluted in distilled water and final volume was made upto 1000 mL.

iii) Methyl red indicator: 0.5% solution in 95% alcohol.

(4) Chloride

i) Potassium chromate indicator solution: 50 g K$_2$CrO$_4$ dissolved in distilled water, silver nitrate solution was added till a definite orange red precipitate appeared. After it was filtered and diluted to 1 L with distilled water.

ii) Standard silver nitrate titrant (0.0141 N): 2.395 g AgNO$_3$ dissolved in distilled water and diluted to 1000 mL.

(5) Sulphate

i) Conditioning reagent: 50 mL glycerol mixed in a solution containing 30 mL concentrated HCl + 300 mL distilled water + 100 mL ethyl alcohol + 75 g NaCl.

(6) Calcium

i) Ammonium purpurate. 150 mg ammonium purpurate dissolved in 100 g ethylene glycol.

ii) EDTA 0.01 M: 3.723 g EDTA dihydrate salt dissolved in distilled water and diluted to 1000 mL.

iii) 1 N NaOH solution: As above in (vi)