APPENDIX 6a

Reagents for serum iron estimation by Ramsay's Dipyridyl method (Varley, 1976).

Reagents

1 22' Dipyridyl, 0.2 percent in acetic acid, 3 percent, v/v.

2 Sodium sulfite, 0.2 M. 2.52 gm of anhydrous salt dissolved in 100 ml water.

3 Chloroform.

4 Standard solution containing 100 mcg iron per ml. Dissolved 0.498 gm of ferrous sulfate, $FeSO_4 \cdot 7H_2O$, in water. Added 1 ml of concentrated sulfuric acid and made to a litre. Alternatively, a solution of ferric ammonium sulfate $(NH_4)_2SO_4 \cdot FeSO_4 \cdot 6H_2O$, containing 0.702 gm per litre can be used.

5 Working standard -
Dilute 3 ml of stock solution to 100 ml with water to obtain a solution containing 3 mcg/ml.

APPENDIX 6b

Reagents for Total Iron Binding Capacity estimation by Ramsay's Dipyridyl method (Varley, 1976).

Reagents

1 Ferric chloride solution, 5 mcg iron per ml in 0.005 N HCl. Prepared a stock solution containing 145 mg of $FeCl_3$ per 100 ml of 0.5 N acid and diluted 1 to 100 ml with water.

2 Magnesium carbonate, "light" for adsorption.

3 Sodium sulfite, 0.2 M, 2.52 gm of anhydrous salt per 100 ml.

4 22' dipyridyl, 0.2 percent in acetic acid, 3 percent, v/v.

5 Chloroform.

6 Standard solution containing 100 mcg iron per ml. Dissolve 0.498 gm of ferrous sulfate $FeSO_4 \cdot 7H_2O$, in water, add 1 ml concentrated sulfuric acid and made to a litre. Alternatively, a solution of ferric ammonium sulfate $(NH_4)_2SO_4 \cdot FeSO_4 \cdot 6H_2O$, containing 0.702 gm per litre can be used.

7 Working standard solution. Dilute 3 ml of stock solution to 100 ml with water to obtain a solution containing 3 mcg per ml.

Standardisation for serum iron.

Serial No.	ml of standard	Concentration of standard (mcg)	ml of water	ml of sulfate	ml of dipyridyl	OD
1	0.2	0.6	1.8	0.5	0.5	0.037
2	0.4	1.2	1.6	0.5	0.5	0.070
3	0.6	1.8	1.4	0.5	0.5	0.116
4	0.8	2.4	1.2	0.5	0.5	0.149
5	1.0	3.0	1.0	0.5	0.5	0.187
6	-	-	2.0	0.5	0.5	-

Depicted graphically in Fig

Standardisation for total iron binding capacity.

Serial No.	ml of standard	Concentration of standard (mcg)	ml of water	ml of sulfite	ml of dipyridyl	OD
1	0.4	1.2	3.6	1.0	1.0	0.035
2	0.8	2.4	3.2	1.0	1.0	0.075
3	1.2	3.6	2.8	1.0	1.0	0.110
4	1.6	4.8	2.4	1.0	1.0	0.149
5	2.0	6.0	2.0	1.0	1.0	0.187
6	-	-	4.0	1.0	1.0	-

Depicted graphically in Fig

Fig21:STANDARD CURVE FOR SERUM IRON

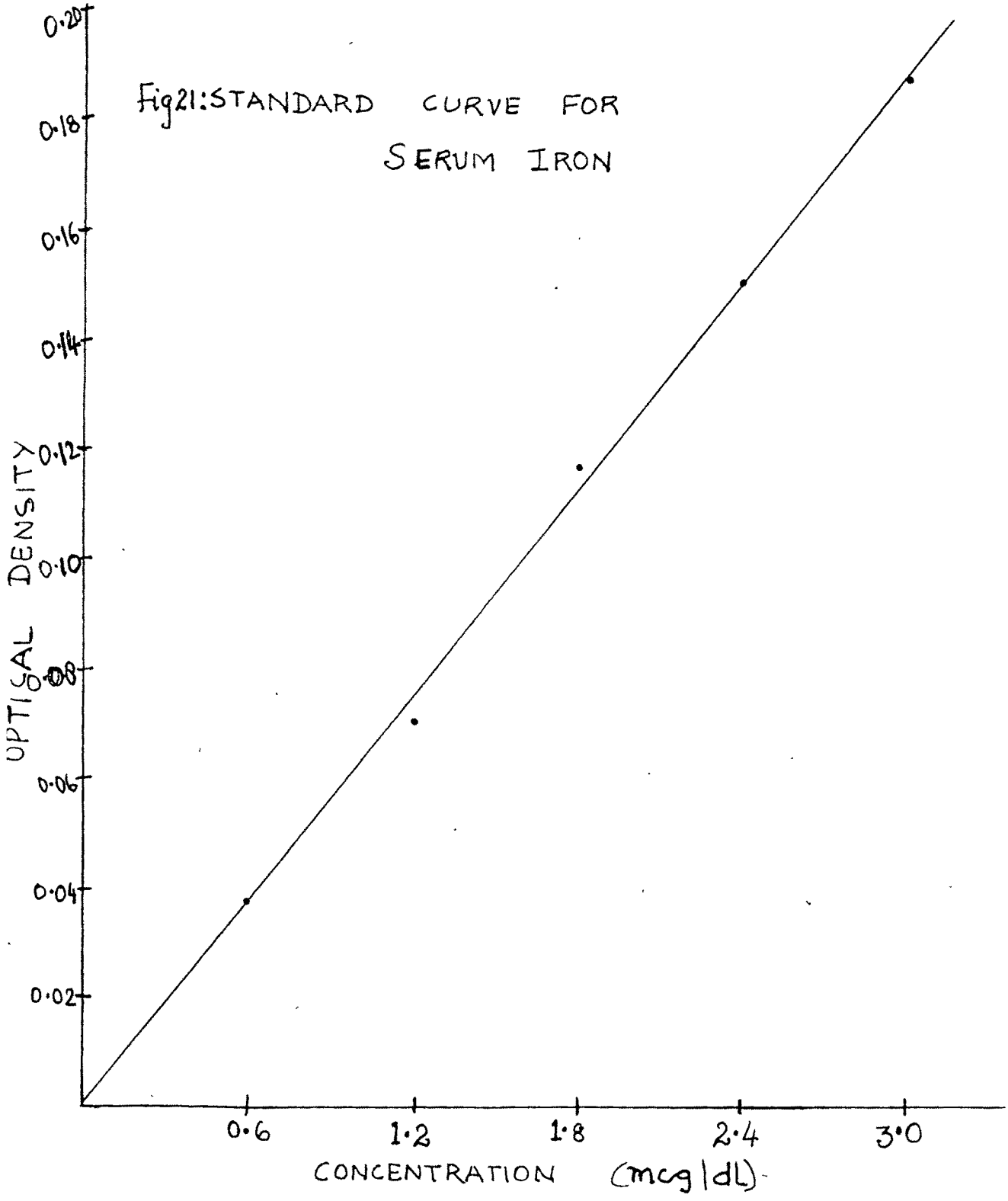

Fig 22: STANDARD CURVE FOR TOTAL IRON BINDING CAPACITY