Summary

The serum vitamin B₁₂ concentrations of patients with pernicious anaemia in relapse rose from low pretreatment levels to within the normal range after single intramuscular injections of 20 to 1,000 µg. of vitamin B₁₂. Concentrations were within the normal range for from 6 to more than 18 days after injections of 20 µg.; from 2 to 22 days after 40 µg.; from 11 to at least 35 days after 80 µg.; from 11 to at least 48 days after 160 µg.; from 39 to 98 days after 320 µg.; and from 34 to at least 58 days after 1,000 µg.

Although the concentrations were within the normal range for these periods, the mean of the concentrations of all the patients in each treatment group fell within a few days of the injections below the mean concentration found in normal subjects.

When the serum vitamin B₁₂ concentrations were normal haemopoiesis was normoblastic; when the concentrations were below the normal range megaloblasts reappeared in the marrow.

Intramuscular injections of 40 µg. of vitamin B₁₂ every 10 days, or of 160 µg. every 21 days, maintained the serum vitamin B₁₂ level of most patients with pernicious anaemia within the normal range. These doses seem to be the minimum requirement for the treatment of most patients with pernicious anaemia in relapse. It is recommended, however, that much larger doses of vitamin B₁₂ (up to 5 mg.) should be given in the first week or two of treatment in order not only to supply enough vitamin B₁₂ for haemopoiesis but also to replenish the greatly depleted tissue reserves.

The serum vitamin B₁₂ concentrations of patients with various megaloblastic anaemias were not altered by treatment with folic acid or folinic acid.

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VITAMIN B₁₂ IN NUTRITIONAL
MACROCYTIC ANAEMIA

BY

C. R. DAS GUPTA, M.B., D.T.M., F.N.I.

J. B. CHATTERJEA, M.D.

Assistant Research Officer, Indian Council of Medical Research

AND

P. BASU, M.B., B.Sc.

Teddington Research Fellow

(From the Department of Haematology, School of Tropical Medicine, Calcutta)

The efficacy of vitamin B₁₂ in pernicious anaemia (P.A.) and its neurological complication is now well established. The exact status of vitamin B₁₂ in the treatment of nutritional macrocytic anaemia (N.M.A.) is, however, still uncertain owing to the scarcity of available reports. Folic acid is universally held to be the drug of choice in N.M.A. The present communication deals with trials of vitamin B₁₂ in 22 cases of N.M.A. undertaken to find out primarily its value in N.M.A., and incidentally to compare its haematonic efficiency with that of folic acid and/or liver extract.

Materials and Methods

The patients were all Indians. Only uncomplicated cases admitted into hospital were chosen for this study. Vitamin B₁₂ was started only when the blood picture had stabilized after admission. In most cases it was administered intramuscularly; in a few cases it was taken orally in tablet form. A course usually lasted for three to five consecutive days. A second and sometimes a third course were given when the improvement was not adequate or sustained—that is, when the blood values either became stationary or dropped after initial improvement. Folic acid was given orally in doses of 30 mg. a day when there was no improvement with vitamin B₁₂ or when the improvement was not sustained. Finally, crude liver extract was given in doses of 3 to 4 ml. a day for six to ten days when the improvement with folic acid was found to lag. Ferrous sulphate, 18 to 21 gr. (1.2 to 1.4 g.) a day was given when hypochromicity was noted after treatment with vitamin B₁₂.

During the whole period the patients had an identical diet consisting of 8 oz. (225 g.) rice, 2 oz. (56 g.) peas, 4 oz. (113 g.) potatoes, 8 oz. green vegetables, 4 oz. bread, 2 oz. butter, 2 oz. sugar, 16 oz. (450 g.) milk, 4 oz. fish, 1 lime, 1 banana. This diet, which is relatively poor in animal protein, has been found to have no haemopoietic activity in N.M.A. A high-protein diet has distinct haemopoietic activity in N.M.A. (Das Gupta and Chatterjea, 1951), and it is therefore imperative that in the evaluation of the efficacy of different haematonic the patients be always on a control diet as above.

The degree of improvement after each course of treatment has been calculated according to the formula of Della Vida and Dyke (1942); for working purposes the improvement rate is indicated as follows:

Very good	=	improvement rate over	90%
Good	=	“ “ between	75 and 90%
Very fair	=	“ “ “	50 “ 74%
Fair	=	“ “ “	30 “ 49%
Tolerable	=	“ “ “	15 “ 29%
Nil	=	“ “ below	15%

Dosage.—Vitamin B₁₂ was not given in graded dosage, the amount usually being determined by the supplies at our disposal. Thus at the beginning, when we had only a limited supply, small doses were used, while during the later part we had liberal supplies and large doses were given.

Bone Marrow.—All cases in the series had cellular megaloblastic bone marrow.

Details of the cases and the results of treatment are shown in Table I.

Vitamin B₁₂ Parenterally

In 21 cases of N.M.A. vitamin B₁₂ was administered intramuscularly, the total amount given to any single case ranging from 30 to 600 µg. in one, two, or three courses of treatment. Six of these cases were also treated orally, five before and one after parenteral therapy. The maximum

improvement in each case after a course of parenteral vitamin B₁₂ is shown in Table II. Though the improvement was very good in ten cases, good in four, and fair in four, it was sustained in four cases only.

A second course of treatment was given in 12 cases—in six (Nos. 1, 3, 6, 7, 10, and 15) the response was increased, in four (Nos. 4, 8, 9, and 13) it was equal, and in two (Nos. 12 and 14) it was decreased compared with the previous course. Two cases (Nos. 10 and 15) had a third course, and in each the response was less than with the previous course.

Correlation of the improvement index with the dose of vitamin B₁₂ is shown in Table III. There was great variation in response in different patients and in the same patient at different times. Strict correlation was not seen between the dose of vitamin B₁₂ and the improvement index, though a better response was usually noted after a large dose—240 µg. or more. Continuation of treatment produced

TABLE I.—Details of Haematological Response at Days Indicated

Case No.	Age and Sex	Haematinic	Route	Dose			Day* of Treatment	Hb (g./%)	R.B.C. (mil./c.mm.)	M.C.V. (m. ³)	Retics.		I.R.†	Remarks
				Daily	No. of Days	Total					%	Day		
1	30 M	Vit. B ₁₂	I.M.	10 µg.	3	30 µg.	{ 0 15 20	3.48 4.93 6.96	1.01 1.23 1.74	148.5 146.3 143.6	10.2	8	Tol. V.G.	Hypochlorhydria. Blood values reached normal without further treatment
		" "	I.M.	20 µg.	2	40 µg.	{ 15 42	10.15 13.48	2.75 4.06	123.6 98.5				
2	25 M	Vit. B ₁₂	I.M.	20 µg.	6	120 µg.	{ 0 15 20	2.03 7.54 7.54	0.52 2.04 2.01	173.0 121.5 124.0	12.8	7	V.G. V.G. V.G.	Hypochlorhydria. Blood values reached normal without further treatment
		Folic acid	Oral	30 mg.	20	600 mg.	{ 15 21	10.44 10.73	3.14 3.21	108.2 102.8				
		Liver ext.	I.M.	4 ml.	6	24 ml.	{ 14	1.89	3.75	96.0				
3	45 M	Vit. B ₁₂	Oral	5 µg.	6	30 µg.	{ 0 15	7.25 7.25	1.66 1.65	120.4 139.3	11.8	6	Nil V.F. V.G.	Isochlorhydria Blood values reached normal level with more vit. B ₁₂
		" "	I.M.	5 µg.	6	30 µg.	{ 15 21	10.44 10.44	2.50 2.48	136.0 133.1				
		" "	I.M.	20 µg.	6	120 µg.	{ 15	12.47	3.77	103.4				
4	28 M	Vit. B ₁₂	I.M.	40 µg.	5	200 µg.	{ 0 15 20 25	3.48 7.54 9.28 9.28	0.73 2.21 2.62 8.29	137.0 117.6 129.7 110.7	7.4	6	V.G. V.G. V.G.	Isochlorhydria
		" "	I.M.	20 µg.	5	100 µg.	{ 10 15	11.60 11.81	3.68 3.95	103.2 101.2				
		Folic acid	Oral	30 mg.	15	450 mg.	{ 15	12.75	4.30	97.6				
5	22 M	Vit. B ₁₂	I.M.	40 µg.	6	240 µg.	{ 0 15 20	2.90 7.83 8.70	0.71 2.34 2.20	154.9 123.9 127.2	11.5	5	V.G. V.G. V.G.	Hypochlorhydria. Left hospital
		Folic acid	Oral	30 mg. 20 mg.	13 8	550 mg.	{ 15 25	10.73 11.02	3.19 3.23	109.7 108.3				
		Liver ext.	I.M.	3 ml.	10	30 ml.	{ 15	12.49	3.73	101.9				
6	60 M	Vit. B ₁₂	I.M.	40 µg.	3	120 µg.	{ 0 15 25	4.93 8.55 8.70	1.24 2.27 2.60	137.1 127.8 107.6	6.3	4	G. V.G. G.	Hypochlorhydria. Left hospital
		" "	I.M.	40 µg. 50 µg.	3 2	220 µg.	{ 15 25	11.02 11.02	3.31 3.12	105.7 112.1				
		Folic acid	Oral	30 mg.	15	450 mg.	{ 7 15	12.18 12.18	3.54 3.58	104.5 103.3				
7	32 F	Vit. B ₁₂	I.M.	40 µg.	3	120 µg.	{ 0 15	5.22 6.67	1.15 1.63	165.2 147.2	12.1	7	F. V.G.	Hypochlorhydria. Blood picture changed to dimorphic type and further improvement was noted after iron and liver extract
		" "	I.M.	40 µg.	3	120 µg.	{ 10	7.83	2.38	126.0				
8	56 F	Vit. B ₁₂	Oral	100 µg.	5	500 µg.	{ 0 7	4.35 5.22	1.15 1.23	139.3 162.5	15.6	6	Nil V.G. V.G.	Isochlorhydria
		" "	I.M.	50 µg.	5	250 µg.	{ 15 21	8.41 9.86	2.49 2.83	128.5 120.1				
		" "	I.M.	50 µg.	6	300 µg.	{ 15	11.60	4.07	95.8				
9	57 M	Vit. B ₁₂	Oral	100 µg.	5	500 µg.	{ 0 7	5.22 5.22	1.36 1.09	125.0 146.7	Nil	7	Nil V.G. V.G.	Hypochlorhydria
		" "	I.M.	50 µg.	5	250 µg.	{ 15	8.41	2.31	125.5				
		" "	I.M.	50 µg.	6	300 µg.	{ 15 30	10.73 13.05	3.22 4.11	111.8 102.1				
10	23 F	Vit. B ₁₂	I.M.	40 µg.	5	200 µg.	{ 0 15	4.93 7.54	1.03 1.86	155.3 145.1	10.1	7	V.F. V.G.	Hypochlorhydria.
		" "	I.M.	20 µg.	5	100 µg.	{ 15 40	10.44 10.73	2.84 3.14	116.1 108.2				
		" "	I.M.	40 µg.	5	200 µg.	{ 10	10.73	2.97	115.4				

TABLE I.—Details of Haematological Response at Days Indicated—(continued)

Case No.	Age and Sex	Haematinic	Route	Dose			Day* of Treatment	Hb (g/%)	R.B.C. (mil./c.mm.)	M.C.V. (m.3)	Retics.		I.R.†	Remarks
				Daily	No. of Days	Total					%	Day		
11	20 M	Vit. B ₁₂	I.M.	20 µg.	5	100 µg.	{ 0 15	2.61 2.90	0.65 0.67	123.0 164.1	7.8	7	Nil G. V.G.	Hypochlorhydria. Blood picture reached normal level without further treatment
		Folic acid	Oral	30 mg.	30	900 mg.	{ 15 25 30	6.38 8.70 8.41	1.87 2.66 2.61	128.3 109.0 106.8	7.6	5		
		Vit. B ₁₂	Oral	15 µg.	6	90 µg.	{ 15 25	10.73 12.32	3.34 3.67	95.8 98.0				
12	47 F	Vit. B ₁₂	I.M.	40 µg.	3	120 µg.	{ 0 15	7.54 8.70	1.68 2.56	136.7 117.1	2.8	7	G. Nil V.F. Nil	Isochlorhydria. Left hospital
		" "	I.M.	40 µg.	3	120 µg.	{ 10	8.12	1.98	131.0				
		Folic acid	Oral	30 mg.	21	630 mg.	{ 15 20	10.15 11.02	2.71 2.87	121.7 118.4				
		Liver ext.	I.M.	3 ml.	6	18 ml.	{ 10	11.02	2.88	121.5				
13	43 F	Vit. B ₁₂	I.M.	40 µg.	3	120 µg.	{ 0 20 30 40	4.06 6.49 10.40 9.86	0.87 2.73 3.12 2.93	172.4 120.8 115.3 109.2	8.3	3	G. G. Nil G.	Isochlorhydria. Left hospital
		" "	I.M.	40 µg.	3	120 µg.	{ 10 20	10.73 11.02	3.26 3.12	98.1 105.7				
		Folic acid	Oral	30 mg.	7	210 mg.	{ 7	10.44	3.26	101.2				
		Liver ext.	I.M.	3 ml.	6	18 ml.	{ 10 30	12.90 13.05	3.60 3.70	102.7 97.2				
		" "	I.M.	40 µg.	3	120 µg.	{ 0 15 25	5.22 8.41 8.12	1.06 2.20 2.23	132.1 127.2 130.0	12.0	4		
		Liver ext.	I.M.	3 ml.	6	18 ml.	{ 15	8.99	2.50	120.0				
14	27 F	Vit. B ₁₂	I.M.	20 µg.	7	140 µg.	{ 0 15 25	5.22 8.41 8.12	1.06 2.20 2.23	132.1 127.2 130.0	12.0	4	G. Tol. Tol. V.G.	Hypochlorhydria. Left hospital
		" "	I.M.	40 µg.	3	120 µg.	{ 10 20	10.73 11.02	3.26 3.12	98.1 105.7				
		Folic acid	Oral	30 mg.	7	210 mg.	{ 7	10.44	3.26	101.2				
		Liver ext.	I.M.	3 ml.	6	18 ml.	{ 10 30	12.90 13.05	3.60 3.70	102.7 97.2				
		" "	I.M.	40 µg.	3	120 µg.	{ 0 15 25	5.22 8.41 8.12	1.06 2.20 2.23	132.1 127.2 130.0	12.0	4		
		Liver ext.	I.M.	3 ml.	6	18 ml.	{ 15	8.99	2.50	120.0				
15	25 F	Vit. B ₁₂	I.M.	40 µg.	5	200 µg.	{ 0 15 25	7.83 8.40 8.70	1.98 2.31 2.97	141.4 138.6 104.3	4.5	4	F. G. Nil V.G.	Isochlorhydria. Left hospital
		" "	I.M.	40 µg.	5	200 µg.	{ 10	8.84	2.84	112.6				
		Folic acid	Oral	30 mg.	15	450 mg.	{ 15 20	8.99 8.99	3.49 3.24	91.6 98.7				
		Liver ext.	I.M.	3 ml.	6	18 ml.	{ 15	9.57	3.58	94.9				
		" "	I.M.	40 µg.	5	200 µg.	{ 10	8.84	2.84	112.6				
		Folic acid	Oral	30 mg.	15	450 mg.	{ 15 20	8.99 8.99	3.49 3.24	91.6 98.7				
16	40 M	Vit. B ₁₂	Oral	10 µg.	4	40 µg.	{ 0 15	10.44 11.6	2.64 3.21	121.2 115.2	3.5	3	V.F. V.F.	Hyperchlorhydria. Left hospital, but further improvement without any other treatment
		" "	Oral	10 µg.	4	40 µg.	{ 15	12.18	3.51	99.7				
17	42 F	Vit. B ₁₂	Oral	15 µg.	3	45 µg.	{ 0 15	9.86 10.15	2.4 2.72	133.3 113.9	Nil		F. Nil F.	Isochlorhydria. Left hospital
		" "	Oral	15 µg.	3	45 µg.	{ 15 20	9.28 10.44	2.80 2.66	103.5 110.8				
		" "	I.M.	15 µg.	5	75 µg.	{ 15	10.44	2.98	110.8				
		" "	I.M.	15 µg.	5	75 µg.	{ 15	10.44	2.98	110.8				
18	18 M	Vit. B ₁₂	I.M.	20 µg.	6	120 µg.	{ 0 15 25 40 50	4.35 5.80 8.41 10.67 8.70	1.14 1.52 2.29 2.85 2.76	131.5 144.7 131.0 112.2 106.0	15.2	7	F. F. Nil V.G.	Absolute achlorhydria at beginning, but isochlorhydria at end of treatment. Left hospital, further improvement without other treatment
		Folic acid	Oral	30 mg.	15	450 mg.	{ 7 15	9.86 9.86	2.87 2.76	104.0 111.0				
		Liver ext.	I.M.	3 ml.	6	18 ml.	{ 7	11.02	3.86	82.5				
		" "	I.M.	40 µg.	6	240 µg.	{ 0 16 25 32	3.19 6.52 6.81 6.67	0.75 1.55 1.85 1.85	160.0 135.6 120.0 122.0	18	10		
		Folic acid	Oral	30 mg.	15	450 mg.	{ 7 15	9.28 9.28	2.18 2.35	131.0 120.5				
		Liver ext.	I.M.	4 ml.	6	24 ml.	{ 15 25	10.44 11.02	2.87 3.45	111.4 101.4				
19	45 M	Vit. B ₁₂	I.M.	20 µg.	6	120 µg.	{ 0 15 25	2.90 4.49 4.64	0.76 1.38 1.26	131.5 123.3 119.0	Nil		F. V.F.	Hyperchlorhydria. Further improvement without other treatment
		Folic acid	Oral	30 mg.	30	900 mg.	{ 15 30	7.54 8.70	2.15 2.97	130.2 101.0				
20	25 M	Vit. B ₁₂	I.M.	20 µg.	6	120 µg.	{ 0 15 25	2.90 4.49 4.64	0.76 1.38 1.26	131.5 123.3 119.0	Nil		F. V.F.	Hyperchlorhydria. Further improvement without other treatment
		Folic acid	Oral	30 mg.	30	900 mg.	{ 15 30	7.54 8.70	2.15 2.97	130.2 101.0				
		" "	I.M.	40 µg.	6	240 µg.	{ 0 15 25	2.90 4.49 4.64	0.76 1.38 1.26	131.5 123.3 119.0	Nil			
21	25 M	Vit. B ₁₂	Oral	15 µg.	5	75 µg.	{ 0 15	2.90 2.46	0.73 0.52	147.9 171.4	4.5	7	Nil Tol. F.	Hypochlorhydria. Improvement continued
		" "	I.M.	15 µg.	5	75 µg.	{ 15	2.32	0.82	146.3	3.1	6		
		Folic acid	Oral	30 mg.	30	900 mg.	{ 15 50	4.35 10.44	1.28 3.47	148.4 100.8				
22	40 F	Vit. B ₁₂	I.M.	40 µg.	3	120 µg.	{ 0 7	5.80 5.51	1.77 1.68	107.3 101.1	Nil		Nil G.	Achlorhydria. Blood picture shows evidence of hypochromicity. Left hospital
		Folic acid	Oral	30 mg.	15	450 mg.	{ 15	6.38	2.54	94.4				

* 0 marks the beginning of the period of observation. † Improvement rate calculated according to the formula of Della Vida and Dyke: V.G.=Very good; G.=good; V.F.=very fair; F.=fair; Tol.=tolerable.

TABLE II.—Result of Parenteral Vitamin B₁₂ Treatment

Improvement Index	No. of Cases	Improvement Sustained		Improvement Not Sustained	
		No.	Case Nos., with Dosage*	No.	Case Nos., with Dosage
Very good ..	10	4	1 (70), 3 (150), 8 (250), 9 (250)	6	2 (120), 4 (300), 5 (240), 6 (340), 7 (240), 10 (500)
Good ..	4	0		4	12 (120), 13 (240), 14 (140), 15 (400)
Fair ..	4	0		4	17 (75), 18 (120), 19 (120), 20 (120)
Tolerable ..	1	0		1	21 (75)
Nil ..	2	0		2	11 (100), 22 (120)

*The amount of vitamin B₁₂ (in µg.) required to produce the response is shown in parentheses after the case number.

TABLE III.—Correlation of Improvement Index With the Dose of Vitamin B₁₂ Given Parenterally

Dose	Very Good	Good	Fair	Tolerable	Nil
Less than 100 µg.	1 (7)	0	1 (17)	1 (21)	0
100 to 200 µg. ..	2 (2, 3)	2 (12, 14)	3 (18, 19, 20)	0	2 (11, 22)
200 to 300 µg. ..	4 (5, 7, 8, 9)	1 (13)	0	0	0
300 to 500 µg. ..	3 (4, 6, 10)	1 (15)	0	0	0
Total ..	10	4	4	1	2

Case numbers are given in parentheses. Cases showing sustained improvement are in italics.

sustained improvement in two cases (Nos. 8 and 9), but did not produce further improvement in three others (Nos. 10, 12, and 14).

Vitamin B₁₂ Orally

Seven cases (Nos. 3, 8, 9, 11, 16, 17, and 21) had vitamin B₁₂ orally. Apart from Case 11 this treatment was given first. The dose was usually small, less than 100 µg., but Cases 8 and 9 each had 500 µg. Of the six cases that were given oral vitamin B₁₂ initially, one (Case 16) showed very fair and sustained improvement with 40 µg., and another (Case 17) showed fair improvement, which was not sustained, with 45 µg. No appreciable improvement was noted in the other four cases, though two of them (Nos. 8 and 9) had large doses. Cases 3, 8, and 17 had normal gastric acidity, Cases 9 and 21 had hypochlorhydria, and Case 16 had hyperchlorhydria. Thus no correlation of the improvement rate was found either with the dose of vitamin B₁₂ given orally or with the state of gastric acidity.

The seventh case (No. 11), with hypochlorhydria, showed very good improvement with 90 µg. of vitamin B₁₂ orally, and the improvement was sustained. Prior to this treatment the patient had shown good improvement with folic acid, which, however, was not sustained. Before the folic acid he had had vitamin B₁₂ parenterally without any appreciable improvement.

Oral and Parenteral Vitamin B₁₂ in Same Patient

Six of the seven cases treated with oral vitamin B₁₂ were also given vitamin B₁₂ parenterally—five after and one before the oral treatment. Of the four cases which did not improve with oral treatment, three (Nos. 3, 8, and 9) showed very good improvement after parenteral treatment, and the improvement was sustained in all of them. In Case 17 the response was about equal with oral and parenteral therapy. Case 11 did not show any response with parenteral treatment, but a very good response with oral treatment, and the improvement was sustained. As mentioned already, this case showed good improvement with folic acid prior to oral treatment. Thus generally a better response was seen with parenteral treatment.

Response to Other Haematinics

Cases showing suboptimal response with vitamin B₁₂ were first given folic acid (14 cases) and those still failing to reach normal level were given liver extract parenterally (eight cases). The improvement index is shown in Table IV.

TABLE IV.—Response to Folic Acid and Liver Extract in Cases Not Showing Sustained Improvement With Vitamin B₁₂

Haematinic	Total No.	Improvement Index			
		Very Good and Good	Very Fair and Fair	Tolerable	Nil
Folic acid ..	14	8 (2, 4, 5, 6, 11, 14, 15, 22)	4 (12, 19, 20, 21)		2 (13, 18)
Liver extract ..	8	4 (2, 5, 13, 18)	2 (15, 19)	1 (14)	1 (12)

The case numbers are given in parentheses.

Compared with parenteral vitamin B₁₂, folic acid produced greater improvement in six cases (Nos. 11, 14, 15, 20, 21, and 22), equal response in four (Nos. 2, 4, 5, and 19), and less response in four (Nos. 6, 12, 13, and 18). The difference was well marked in four cases—in two (Nos. 11 and 22) there was no improvement with vitamin B₁₂, but good improvement with folic acid, while the other two (Nos. 13 and 18) showed good and fair improvement with vitamin B₁₂, but no improvement with folic acid.

Compared with vitamin B₁₂ and folic acid, liver extract produced definitely better response in one case (No. 18), about equal response in three cases (Nos. 2, 5, and 19), and less response in three cases (Nos. 12, 14, and 15). One case (No. 13) showed about the same response as with vitamin B₁₂, but a better response than with folic acid. The difference in response was well marked in three cases, being in favour of liver extract in Case 18, and in favour of vitamin B₁₂ and folic acid in Cases 12 and 14.

Reticulocytosis.—Adequate reticulocyte response was not seen in any of the cases after vitamin B₁₂ orally or parenterally, even when the improvement in the red-cell count was very good (Table I).

Macrocytosis.—In all except two cases (Nos. 1 and 11) macrocytosis persisted for a considerable period after vitamin B₁₂ therapy, when the increase in the red-cell count was found to be very good, including those cases in which the blood reached normal levels without any further treatment.

Discussion

Published reports on the effect of vitamin B₁₂ in N.M.A. are still too scanty to permit any definite conclusion to be drawn. Patel (1948), Spies *et al.* (1949), and Chaudhuri (1951) have reported favourably on the effect of vitamin B₁₂. On the other hand, contrary views have been expressed by Luhby (1949), Bethell (1950), and Davidson and Innes (1951). Our earlier impression had been that vitamin B₁₂ produces a suboptimal response in N.M.A. (Das Gupta, 1951, Das Gupta and Chatterjea, 1951).

In the present series initially good improvement was noted in 14 out of the 22 cases (64%), but the improvement was sustained and reached the normal level with vitamin B₁₂ alone in only five cases (23%). Reticulocytosis after vitamin B₁₂ was poor in all cases, and there was no correlation between the improvement index and the reticulocyte response. After vitamin B₁₂ macrocytosis was found to persist for a long time in most of the cases which showed very good improvement. Folic acid was given after vitamin B₁₂ in 14 cases, with good response in eight of them, but the blood picture reached a normal level in only four. Still later, crude liver extract was given to eight cases which had failed to reach a normal level after vitamin B₁₂ and folic acid, and all except one showed further response, the blood picture reaching a normal level in three of them.

In general the response to vitamin B₁₂ had no distinct correlation with the dose of vitamin B₁₂, with the route of administration, or with the state of gastric acidity, but the maximum response was usually obtained with 200 to 300 µg. of vitamin B₁₂, given intramuscularly. Great variation in response to vitamin B₁₂ was noted not only among the different cases but also in the same case at different times. In some cases large doses of vitamin B₁₂ failed to produce

any further improvement after good initial improvement had been obtained with smaller doses. All these cases, however, showed further improvement with folic acid.

The present study indicates that both folic acid and vitamin B₁₂ are valuable agents in the treatment of N.M.A., and we are inclined to think that N.M.A. represents deficiency of both folic acid and vitamin B₁₂. Deficiency of folic acid is probably the major and primary factor, as is evident from the good response obtained with folic acid in cases of N.M.A. (Das Gupta and Chatterjea, 1946), and of macrocytic anaemia in pregnancy (Das Gupta and Chatterjea, 1949), and deficiency of vitamin B₁₂ the minor and secondary factor.

In an average case of N.M.A., administration of vitamin B₁₂ will correct only the B₁₂ deficiency and produce a response which might appear to be optimum initially. During the period of B₁₂ activity some amount of available folic acid will be used up, producing further deficiency of folic acid in the system. In this way extreme deficiency of folic acid may result, and at this stage no further improvement is possible with vitamin B₁₂. Obviously folic acid in this situation will in the majority of cases bring the blood picture to normal. The complete success of vitamin B₁₂ in an occasional case may be explained by assuming that a considerable amount of folic acid is available in such a case for utilization by B₁₂.

Failure of oral vitamin B₁₂ therapy even with massive dosage, and subsequent improvement with smaller dosage of the vitamin given parenterally, proves that absorption of B₁₂ must be severely impaired in N.M.A. In N.M.A. the degree of this malabsorption may not run parallel with the gastric acidity, and may in fact be quite independent of gastric hydrochloric acid. It is probable that in N.M.A. the absorption of vitamin B₁₂ is determined by the condition of intestinal absorptive surface. We have observed a temporary and reversible type of histamine-fast achlorhydria in N.M.A. In these cases (No. 18 of the present series is an example), along with the improvement of anaemia there is a return of hydrochloric acid. It is possible that a few of these cases may also have a temporary phase of diminished intrinsic factor activity.

In the majority of cases of N.M.A. folic acid alone would be sufficient, because vitamin B₁₂ deficiency is a minor one and never extreme. As the requirement of vitamin B₁₂ is very small this is easily obtainable from the diet, especially after administration of folic acid, which would improve the absorption of B₁₂ from the gastro-intestinal tract. This will explain the success we obtained with folic acid in cases of N.M.A. and the macrocytic anaemia of pregnancy.

It should, however, be emphasized that N.M.A. cases in different parts of the world may vary in their degree of deficiency of vitamin B₁₂ and folic acid, depending on the dietary habits and gastro-intestinal condition. The response to vitamin B₁₂ and/or folic acid depends both on absolute and on relative deficiency of the two factors, which may explain the difference in response obtained by workers in different places.

The superiority of crude liver extract over folic acid and vitamin B₁₂ in some cases of N.M.A. may be explained by its complex character, its content of vitamin B₁₂, folic acid, folinic acid, and other members of the vitamin-B complex possibly having a synergistic effect, ensuring optimum haemopoiesis. Whether or not liver extract contains any other still unidentified factor—for example, accessory factors of Jacobson and Subbarow (1937) or protein synthesis factor of Owren (1951)—which may be valuable in the treatment of N.M.A., cannot be stated with certainty in the present state of our knowledge.

Summary

The results of treatment with vitamin B₁₂ administered intramuscularly in 21 cases and orally in seven cases of nutritional macrocytic anaemia are reported.

Though the initial improvement index was good in 64% of cases, sustained remission was obtained in only 23%. Reticulocytosis after vitamin B₁₂ therapy was inadequate, and there was no correlation between the improvement index and the reticulocyte response. Macrocytosis tended to persist even in cases showing very good improvement. Folic acid and crude liver extract evoked adequate or better response in most cases not showing sustained remission with vitamin B₁₂.

In general, maximum response was obtained with 200–300 µg. of vitamin B₁₂ given intramuscularly, but this response had no distinct correlation with the dose. The response to oral vitamin B₁₂ also had no correlation either with the dose or with the state of gastric acidity.

The present studies definitely indicate that N.M.A. represents deficiency of both folic acid and vitamin B₁₂, deficiency of folic acid being the major and primary factor, and that of vitamin B₁₂ the minor and secondary factor in most cases.

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In his last annual report on the health of Edinburgh (*Annual Report of the Public Health Department for the Year 1952*) Dr. W. G. Clark, who has recently retired from the post of medical officer of health, has included a chapter on tuberculosis. After reminding us that it was in Edinburgh that the tuberculosis dispensary system was first conceived—a development which was to have a profound effect on tuberculosis practice throughout the world—Dr. Clark examines the recent trend of the disease in the city. While the death rate fell from 60 deaths per 100,000 of the population immediately before the war and 62 in 1948 to 26 in 1952, the notified incidence of the disease rose steadily; in 1939 there were 92 notifications per 100,000, in 1952 there were 154. These increases are reflected in the number of cases of pulmonary tuberculosis on the Tuberculosis Register; in 1938 their number was 1,769, by 1952 it was 3,524. The easy explanation for the rise is that it was due to improved case-finding as a result of the widespread adoption of mass miniature radiography. However, this view was strongly challenged by the Tuberculosis Committee of the Scottish Health Services Council in 1951. Now Dr. Clark, on the basis of more up-to-date information, is able to show that at least a sizable proportion of the increase in notifications was due to earlier case-finding. He adds: "It would be unwise, however, to accept this as a complete answer."