

Serum Vitamin B₁₂ Levels and Vitamin B₁₂ Binding Capacity in Pregnant and Non-pregnant Europeans and West Indians

T. S. LOW-BEER,* B.M., B.CH., M.R.C.P.; C. F. MCCARTHY,* M.D., M.R.C.P., M.R.C.P.I.
W. I. AUSTAD,* M.B.(N.Z.), M.R.C.P.; A. BRZECHWA-AJDUKIEWICZ,* M.B., CH.B., B.SC.
A. E. READ,* M.D., F.R.C.P.

Brit. med. J., 1968, 4, 160-161

Summary: The vitamin B₁₂ level and the capacity of serum to bind B₁₂ are higher in the West Indian population living in Great Britain than in Europeans. The B₁₂ level fell during pregnancy in both groups but remained higher in the West Indians. West Indians had higher levels of IgG.

Introduction

During a survey of the effects of physiological doses of folic acid in pregnancy it was observed that the serum vitamin B₁₂ level seemed to be higher in West Indians than in Europeans. This paper presents these results and further studies of serum B₁₂ levels and serum B₁₂ binding capacities in non-pregnant West Indian and European subjects.

Patients and Methods

The patients studied initially were those attending for antenatal care at a Bristol health centre. Non-pregnant West Indian patients attending a Bristol general practitioner, hospital staff, and medical students were also studied.

Venous blood was taken at the first visit to the centre for estimation of haemoglobin and serum iron, B₁₂, and folate levels. At each subsequent monthly visit blood was taken for haemoglobin and folate estimation. The patients were admitted to hospital for delivery, and these investigations were repeated. A dietary history was taken at the first and the last antenatal visit, from which the B₁₂ content of the diets was estimated, the tables of McCance and Widdowson (1960) being used. Serum folate levels were measured by microbiological assay (*Lactobacillus casei* ATCC 746), the normal range being 5.9 to 21 µg./ml. Serum B₁₂ levels were estimated by microbiological assay, the normal range being 140 to 900 µg./ml. In the non-pregnant group serum bilirubin, alkaline phosphatase, serum proteins, and paper electrophoresis were carried out. Vitamin B₁₂ binding was estimated by the method of Gottlieb *et al.* (1965). Quantitative immunoelectrophoresis was carried out by the method of Mancini *et al.* (1965) with immunoplates and standard sera (Hyland Inc.). Statistical tests of significance were carried out by use of Student's *t* test, with logarithmic transformation of data where needed to normalize the distribution.

Results

Non-pregnant Patients.—The results of serum B₁₂ and unbound B₁₂ capacity are given in Table I. The serum B₁₂ levels and the unbound B₁₂ binding capacity were higher in West Indians than in the Europeans. The highest five values of unbound B₁₂ binding capacity do not fall within the normal distribution. If these are excluded the mean falls, but is still

* Department of Medicine, University of Bristol at the Royal Infirmary and Southmead Hospitals.

TABLE I.—Males and Non-pregnant Females*

		West Indians	European	P
Vitamin B ₁₂ (µg./ml.)	Males	Mean 687 ± 231 (11) Range 375-1,130	381 ± 28.9 (9) 260-525	< 0.01
	Females	Mean 781 ± 319 (31) Range 370-1,500	391 ± 88.2 (9) 250-530	< 0.01
Unbound B ₁₂ binding capacity (µg./ml.)	Males	Mean 1,610 ± 444 (11) Range 1,037-2,437	855 ± 257 (9) 544-1,283	< 0.025
	Females	Mean 1,832 ± 1,120 (31) Range 720-5,131	1,164 ± 384 (9) 611-1,794	< 0.02

* Mean results ± 1 S.D. No. of subjects in parenthesis.

significantly greater than the European level ($P < 0.02$). The haemoglobin levels were similar in both groups. In all 30 West Indians tested, serum bilirubin, alkaline phosphatase, serum albumin, and paper electrophoresis were within normal limits. The immune globulin levels in the West Indians are given in Table II. The mean level of IgG is much higher in the West Indians than in Europeans for this laboratory (1,200 ± 200). The IgA level is higher in the males than in the females ($P < 0.005$), but does not differ significantly from normal IgA

TABLE II

Immune globulins*			
Males		Females	
IgA mg./100 ml.	350 ± 155 (10)	215 ± 88 (20)
IgG mg./100 ml.	2,000 ± 133 (10)	2,200 ± 533 (20)
IgM mg./100 ml.	96.5 ± 44 (10)	70.9 ± 28.1 (20)

* Mean results ± 1 S.D. No. of subjects in parenthesis.

levels in Europeans. The B₁₂ levels in the non-pregnant group are higher than the levels in the group during or immediately after delivery (West Indian $P < 0.005$; European $P < 0.025$).

Pregnant Patients.—The details are given in Table III. The B₁₂ levels in the West Indians in early pregnancy and immediately after delivery are higher than in the Europeans at a similar stage of pregnancy ($P < 0.001$). In each group the level of serum B₁₂ is lower after delivery than in early pregnancy ($P < 0.001$). The levels of haemoglobin and serum folate and the dietary intake of B₁₂ are higher in the Europeans than in the West Indians.

TABLE III.—During Pregnancy and Immediately After Delivery*

		West Indian	European	P
Vitamin B ₁₂ ³ (µg./ml.)	Early pregnancy	Mean 649 ± 219 (39) Range 275-1,360	308 ± 101 (142) 129-900	< 0.001
	After delivery	Mean 572 ± 156 (12) Range 175-640	257 ± 85 (35) 105-390	< 0.001
Haemoglobin (g./100 ml.)	..	12.1 ± 1.94 (38)	12.6 ± 1.01 (141)	< 0.005
Serum folate (mµg./ml.)	..	3.13 ± 1.60 (37)	4.33 ± 2.50 (131)	< 0.001
Vitamin B ₁₂ intake (mµg./day)	..	4.98 ± 1.29 (39)	5.66 ± 1.55 (137)	< 0.01

* Mean values ± 1 S.D. No. of subjects in parenthesis.

Discussion

No reason is apparent for the higher B₁₂ levels and the higher binding levels observed in the West Indians. It is not related to a dietary intake of B₁₂, as the intake of B₁₂ is lower

in the West Indians. Neither can they be attributed to liver disease, in which the B₁₂ level may be raised.

The occurrence of a raised IgG in the West Indians suggests that the primary abnormality may be one of protein formation. It is not likely that the higher B₁₂ levels are due to its binding directly to IgG, because neither of the two B₁₂ binding proteins (Hall and Finkler, 1963, 1965) is a gammaglobulin. Transcobalamin I is an α 1-globulin and transcobalamin II elutes from diethylaminoethanol cellulose just following the majority of γ - and β -globulins. High levels of IgG have been reported in Gambians (Rowe *et al.*, 1968), in the Congolese (Michaux, 1966), and in Nigerians (Turner and Voller, 1966). Rowe and his colleagues think that this difference may be genetic in origin. Liver disease and recurrent infections are less likely alternatives. High B₁₂ levels and high levels of B₁₂ binding capacity have recently been reported in Southern Nigerians by Fleming (1968). The mean serum B₁₂ \pm 1 S.D. was 575 \pm 16 μ g./ml.; the mean unbound serum B₁₂ binding capacity was 1,802 μ g./ml. with a range of 1,433 to 2,083 μ g. Brandt and Metz (1961) and Brandt *et al.* (1963) reported high serum B₁₂ levels in some of the Bantu population.

All the West Indians studied here were of African descent. Their high levels of B₁₂ may be an inherited feature which is independent of the nutritional and environmental changes of their living in the West Indies and Great Britain. Similar considerations apply to the high level of IgG.

Whether the high B₁₂ level is of any importance or value to the West Indians is speculative. In some conditions, particularly liver abscess, the finding of high B₁₂ levels is of help in arriving at the correct diagnosis (Neale *et al.*, 1966). Clearly a high B₁₂ level in a West Indian patient suspected of having a liver abscess could be misleading. If the total body stores of B₁₂ are increased in West Indians this might afford some protection against a development of B₁₂ deficiency.

Lowering of the B₁₂ in pregnancy has previously been reported (Heinrich, 1954; Spray and Witts, 1958; Lowenstein *et al.*, 1960). Clearly West Indians are not exceptions, as the fall seen in this study was similar in the Europeans and the West Indians. The B₁₂ level reaches the lowest values at the end of pregnancy. The progressive fall in the level is thought to be due to the concentration of the vitamin across the placenta by the foetus (Heinrich, 1954; Killander and Vahlquist, 1954).

We thank Dr. C. Walker for the immunoglobulin estimations and for his help in the preparation of this paper, and Drs. M. and K. Burke, Dr. R. Pardoe, and the medical, nursing, and administrative staff at the Charlotte Keele Clinic for considerable help with this study. We gratefully acknowledge the generous financial and other assistance from Riker Ltd.

REFERENCES

- Brandt, V., and Metz, J. (1961). *S. Afr. J. med. Sci.*, **26**, 1.
 Brandt, V., Kerrich, J. E., and Metz, J. (1963). *S. Afr. J. med. Sci.*, **28**, 125.
 Fleming, A. F. (1968). *W. Afr. med. J.*, **17**, 25.
 Gottlieb, C., Kam-Seng Lau, Wasserman, L. R., and Herbert, V. (1965). *Blood*, **25**, 875.
 Hall, C. A., and Finkler, A. E. (1963). *Biochem. biophys. Acta (Amst.)*, **78**, 234.
 Hall, C. A., and Finkler, A. E. (1965). *J. Lab. clin. Med.*, **65**, 459.
 Heinrich, H. C. (1954). *Klin. Wschr.*, **32**, 205.
 Killander, A., and Vahlquist, B. (1954). *Nord. Med.*, **51**, 777.
 Lowenstein, L., Lalonde, M., Deschenes, E. B., and Shapiro, L. (1960). *Amer. J. clin. Nutr.*, **8**, 265.
 McCance, R. A., and Widdowson, E. M. (1960). *Spec. Rep. Ser. med. Res. Coun. (Lond.)*, No. 297.
 Mancini, G., Carbonara, A. O., and Heremans, J. F. (1965). *Immunochemistry*, **2**, 235.
 Michaux, J.-L. (1966). *Ann. Soc. belge Méd. trop.*, **46**, 483.
 Neale, G., Caughey, D. E., Mollin, D. L., and Booth, C. C. (1966). *Brit. med. J.*, **1**, 382.
 Rowe, D. S., McGregor, I. A., South, S. J., Hall, P., and Williams, K. (1968). *Clin. exp. Immunol.*, **3**, 63.
 Spray, G. H., and Witts, L. J. (1958). *Brit. med. J.*, **1**, 295.
 Turner, M. W., and Voller, A. (1966). *J. trop. Med. Hyg.*, **69**, 99.

Medical Memoranda

Apparent Cure of Cancer with Cyclophosphamide

Brit. med. J., 1968, **4**, 161-162

Cure of cancer of the upper oesophagus by cytotoxic drugs is a rare event. A small number of two-year survivors of cancer of the head and neck by direct intra-arterial injection of alkylating agents are reported by Harrison (1964). Rundles (1962) quoted the case of a woman with an oesophageal tumour who lived two years after cyclophosphamide therapy.

CASE REPORT

A 58-year-old man presented on 10 March 1962 with a seven-weeks history of hoarseness and pain, difficulty in swallowing, and a swelling in the neck. He had also lost 2 st. (12.7 kg.) in weight.

On examination he was thin, emaciated, pale, and anxious. There was a hard fixed mass in the left supraclavicular fossa, about 3½ in. (9 cm.) in diameter. This felt like a mass of malignant lymph nodes. A barium swallow showed a smooth stricture at the upper end of the oesophagus.

On 16 March oesophagoscopy revealed a smooth, hard stricture at 18 cm. from the upper gums, and bronchoscopy at the same time showed a right vocal cord palsy and a granular tumour projecting from the back wall of the trachea 2.5 cm. below the vocal cords.

A biopsy specimen of this mass was taken and reported on as follows: "The specimen is a portion of tissue covered by squamous epithelium, which is not ulcerated in the tissue received. Beneath the epithelium there is widespread infiltration by a carcinoma composed of large pleomorphic cells, with many mitotic figures. There is heavy infiltration by polymorphs, and the neoplasm shows no evidence of differentiation. The findings are those of infiltration by carcinoma compatible with a primary origin in the oesophagus or the trachea."

No treatment was given, but nearly a month later the patient was readmitted with nearly absolute dysphagia, and a further oesophagoscopy was carried out on 14 April. The malignant stricture was now much tighter and it was dilated, and a No. 12 Mousseau-Barbin tube cut off to a suitable length was passed through the stricture and left in position. On 17 April he was given 1 g. of Endoxana (cyclophosphamide) diluted in 50 ml. of water intravenously. He had a sharp reaction with nausea and vomiting and leucopenia. He was so ill that it seemed he would very soon die, and accordingly he was allowed to go home.

On 10 August he reappeared saying that swallowing liquids caused him to cough, but he had no difficulty with solids. This statement was surprising in the circumstances, but even more remarkable was the fact that the mass in the left side of the neck had entirely disappeared, and moreover he looked remarkably well and had gained a lot of weight.

Oesophagoscopy on 15 August revealed no sign of the Mousseau-Barbin tube or of the malignant stricture. In its place were two clean punched-out holes through into the trachea, each about 5 mm. in diameter. Tracheoscopy still showed the right vocal cord palsy and the oesophagotracheal fistula.