Comparison of the effects of food and carbachol on the absorption of vitamin B₁₂ after partial gastrectomy

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SUMMARY Absorption of radioactive vitamin B₁₂ was measured by the Schilling procedure in 14 patients who had had a partial gastrectomy. Injection of carbachol (250 µg) before the administration of the test dose increased absorption to a much smaller degree than the ingestion of a light meal containing a minimal quantity of the natural vitamin. The results support the belief that food may improve absorption of vitamin B₁₂ after partial gastrectomy by stimulating the secretion of a small residual amount of intrinsic factor.

After partial gastrectomy absorption of vitamin B₁₂ is often impaired by reduction in secretion of Castle's intrinsic factor (Brodine, Friedman, Saenger, and Will, 1959; Lous and Schwartz, 1959; Mollin and Hines, 1964). In some patients absorption is improved if the test dose of radioactive vitamin B₁₂ is given either with a meal containing only a minimal quantity of the natural vitamin or after an injection of histamine (Deller, Germar, and Witts, 1961; Turnbull, 1967), and it has been suggested that this improvement is due to stimulating secretion of a small amount of intrinsic factor. Carbachol may improve absorption of vitamin B₁₂ in some patients with atrophic gastritis (Whiteside, Mollin, Coghill, Williams, and Anderson, 1964), presumably by some effect on intestinal motility or vascularity since it does not stimulate the secretion of intrinsic factor (Ardeman, Chanarin, and Doyle, 1964; Ardeman and Chanarin, 1965). The possibility that food might similarly affect absorption was therefore investigated.

METHODS

Absorption of radioactive vitamin B₁₂ was measured by the Schilling procedure (Schilling, Clatanoff, and Korst, 1955) in 14 patients who had undergone partial gastrectomy from six to 24 years previously. They fall into two groups. The first seven patients (group A) had previously had a series of measurements of absorption of vitamin B₁₂ as follows: (1) the standard test, in which the radioactive vitamin B₁₂ was given alone; (2) the meal test, in which it was given after the patient had eaten a vegetable risotto containing a minimum of natural vitamin B₁₂; (3) the augmented test in which it was given 30 minutes after an injection of histamine (0.04

mg/kg); and (4) in some cases a test in which it was given with intrinsic factor concentrate. The techniques used and the results of these tests have been described previously (Turnbull, 1967). In these patients a further test was carried out in which the radioactive vitamin B₁₂ was given 10 minutes after the subcutaneous injection of 0.25 mg carbachol with the patient fasting. The interval between the carbachol test and the previous tests in these patients varied between two and 15 months (average eight months).

Since the small effect of the carbachol injection in these patients might have been due to a further reduction in secretion of intrinsic factor by the stomach remnant in the interval which had elapsed since the previous tests, another seven subjects were studied (group B) and in these the order of the tests varied. The standard test was always done first. Alternate patients then had the carbachol test next and the meal test third and *vice versa*. A test with intrinsic factor was done last. Each test was performed not less than seven days following an injection of vitamin B₁₂. In six of the subjects in group B the standard, meal, and carbachol tests were all carried out within 14 weeks. In one the tests were spread over five months because one test had to be repeated following the use of a faulty batch of radioactive vitamin B₁₂.

The subjects in group B were selected by the same criteria as those in group A: namely, patients who had been treated recently for anaemia which was considered to be a sequel to partial gastrectomy and to be uncomplicated by other disease; who were judged to be sufficiently reliable to make accurate 24-hour collections of urine at home; who had been found to have some impairment of absorption of vitamin B₁₂ in a standard Schilling test.

As in the previous studies (Turnbull, 1967) the oral dose of radioactive vitamin B₁₂ was 1 µg (approximately 1 µCi ⁵⁸Co or occasionally ⁵⁷Co cyanocobalamin) administered in 20 ml water; the 'flushing' dose of cold

cyanocobalamin was 1,000 µg given intramuscularly one hour after the oral dose. Except in the meal tests, only an early morning cup of tea was allowed before the oral dose was given, and no food or drink for two hours after. A light, uncooked breakfast was allowed three or more hours before the meal test. The radioactivity in a 24-hour collection of urine begun immediately after the injection of the 'flushing' dose was compared to that of a suitably diluted standard (1/10th of the oral dose) using a 16-tube Geiger-Müller ring counter (Veall and Vetter, 1952). The effect of intrinsic factor on absorption in patients in group B was evaluated by giving the radioactive vitamin B₁₂ with 50 mg of vitamin B₁₂-free intrinsic factor concentrate (A. Revai and Co (Chemicals) Ltd); this dose will restore to normal the absorption of vitamin B₁₂ in patients with Addisonian pernicious anaemia.

RESULTS

The results of the tests of absorption of radioactive vitamin B₁₂ in both groups of patients are shown in Table I. The significance of the differences between absorption in the standard and modified tests was assessed by comparison with the results of duplicate standard tests in 14 postgastrectomy patients by analysis of variance (Moroney, 1956) as previously reported (Turnbull, 1967).

In group A the mean excretion of radioactivity in the carbachol tests (6.5%, SEM ± 0.8%) was

slightly greater than in the standard tests (5.2% ± 0.6%), but this difference was not significant ($P > 0.1$). Excretion in the meal tests (mean 11.7% ± 1.2%) was significantly greater than that in both the standard tests ($P < 0.001$) and the carbachol tests ($P < 0.001$).

In group B mean excretion in the carbachol tests (6.4% ± 0.9%) was greater than in the standard tests (3.9% ± 0.6%) and this difference was probably significant ($0.05 > P > 0.025$). Excretion in the meal tests (mean 10.3% ± 1.3%) was again significantly greater than that in the standard tests ($P < 0.001$) and that in the carbachol tests ($0.025 > P > 0.01$).

If the observations in both groups are combined, mean excretion in the carbachol tests (6.5% ± 0.6%) was significantly greater than in the standard tests (mean 4.6% ± 0.5%) ($0.025 > P > 0.01$). Excretion in the meal tests (11.0% ± 0.9%) was significantly greater than in the standard tests ($P < 0.001$), and was again significantly greater than that in the carbachol tests ($P < 0.001$). Excretion of radioactivity was greater when the radioactive vitamin B₁₂ was given with hog intrinsic factor concentrate in all 10 patients in whom it was possible to carry out this extra test.

DISCUSSION

Carbachol has been shown to increase absorption of radioactive vitamin B₁₂ in some patients with atrophic gastritis but not in those with pernicious anaemia (Whiteside *et al*, 1964). No apparent effect on absorption was seen in eight patients after partial gastrectomy studied by Dellipiani and Seaton (1965), but little would have been expected in four of these in whom absorption was normal in the standard Schilling test. In the present studies absorption of radioactive vitamin B₁₂ appeared to be slightly improved by the injection of carbachol, but the improvement only attained probable statistical significance in group B when the figures were compared with those of duplicate standard tests in 14 postgastrectomy patients. The smaller effect of carbachol in the patients in group A may be attributable to some further deterioration in intrinsic factor secretion by the stomach remnant in the interval between the tests. In both groups a much larger and statistically more significant enhancement of absorption was seen when the test dose of radioactive vitamin B₁₂ was given with a meal containing a minimal quantity of natural B₁₂. Injection of histamine (0.04 mg/kg) has similarly enhanced absorption in many patients who have had a partial gastrectomy, including all seven patients in group A (Turnbull, 1967).

TABLE I
TWENTY-FOUR HOUR URINARY RADIOACTIVITY
IN SCHILLING TESTS
Radioactivity in Schilling Test (%)

Patient	Standard	Meal	Carbachol	Intrinsic Factor Added
Group A				
W.M.	5.5, 2.2, 3.5	9.7	4.5	10.0
W.B.	3.8, 5.4	11.0	6.1	15.3
D.B.	2.7, 6.6	10.5	5.9	—
N.S.	4.4, 5.3	13.6	4.9	—
S.T.	6.7	12.4	7.1	9.9
O.M.	8.2	18.7	11.1	—
T.D.	4.4, 2.4	6.2	6.1	—
Mean ± SEM	5.2 ^a ± 0.6	11.7 ^{a,b} ± 1.2	6.5 ^b ± 0.8	—
Group B				
C.B.	1.7, 3.6	6.1	9.7	12.1
E.L.	4.5	14.1	6.3	9.3
F.B.	6.8	13.6	4.8	10.8
J.R.	2.3	6.8	4.5	11.8
J.N.	2.6	8.4	3.6	8.7
N.R.	4.5	13.0	8.3	9.1
D.S.	4.2	10.3	7.9	15.7
Mean ± SEM	3.9 ^{c,d} ± 0.6	10.3 ^{c,e} ± 1.3	6.4 ^{d,e} ± 0.9	—
Groups A and B				
Mean ± SEM	4.6 ^{f,g} ± 0.5	11.0 ^{f,h} ± 0.6	6.5 ^{g,h} ± 0.6	—
a-a, b-b, c-c, f-f, h-h Significant difference $P < 0.001$				
e-e, g-g Significant differences $0.025 > P > 0.01$				
d-d Probable significant differences $0.05 > P > 0.025$				

Malabsorption of vitamin B₁₂ after partial gastrectomy is usually due to inadequate secretion of intrinsic factor (Brodine *et al*, 1959; Lous and Schwartz, 1959; Mollin and Hines, 1964). The improvement in absorption in these patients when the test dose of vitamin B₁₂ is given with a 'B₁₂-free' meal or after histamine may most easily be attributed to the stimulation of a small residual secretion of intrinsic factor. In patients with Addisonian pernicious anaemia the 'B₁₂-free' meal does not alter the absorption of radioactive vitamin B₁₂ even when it is given with a suboptimal amount of intrinsic factor concentrate (Deller *et al*, 1961; Turnbull, 1967), thus making unlikely an alternative hypothesis that the meal might improve absorption after partial gastrectomy by protecting a small residual secretion of intrinsic factor from destruction in the intestinal lumen.

A third possibility would be that the meal improves absorption by altering either intestinal motility or vascularity. This is the probable explanation for the enhancement of absorption of vitamin B₁₂ by carbachol in patients with atrophic gastritis (Whiteside *et al*, 1964), since this drug does not stimulate secretion of intrinsic factor (Ardeman *et al*, 1964; Ardeman and Chanarin, 1965). The observations presented here show that carbachol may cause a marginal improvement in absorption of vitamin B₁₂ in patients in whom the 'B₁₂-free' meal has a marked effect. The findings make it unlikely that the meal exerts its effect on absorption solely by altering motility and/or vascularity, and support

the conclusion that it stimulates the secretion of a small amount of intrinsic factor.

It is a pleasure to thank the staff of the Dietetic Department for the meals, Revai Chemicals for the supply of intrinsic factor concentrate, Dr C. R. B. Joyce for advice on the statistical analysis, and Miss L. Frazer for technical assistance.

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