

VITAMIN B₁₂ ABSORPTION STUDIES USING RADIOACTIVE VITAMIN B₁₂ : COMPARISON OF WHOLE BODY RETENTION AND URINARY EXCRETION AFTER FLUSHING DOSE (SCHILLING'S TEST)

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Summary: Vitamin B-12 absorption of 12 healthy young male subjects was determined by using labelled vitamin B-12 by Schilling's test and whole body retention test. The whole body retention test was performed first using low energy Co57 isotope and a week later, Schilling's test was performed. Whole body retention showed a mean absorption of 56.5% while Schilling's test showed mean absorption of 16.9% in the same subjects. Data supports the whole body retention test being more accurate.

Key words: whole body counting Schilling's test vitamin B-12 absorption

INTRODUCTION

Schilling (8) introduced oral administration of radio labelled vitamin B-12 and measurement of urinary radioactivity (after a flushing dose of non radioactive vitamin B-12 intramuscularly) as a means of assessing the absorption of orally administered vitamin B-12 (8). Since then, several workers have used other parameters like whole body retention (1), hepatic radioactivity measurements (5), 8 hour plasma radioactivity measurement (3), and measurement of foecal radioactivity (6). Variable estimates were obtained by different workers using different methods, but Schilling's test is widely used and more recently whole body retention test is being favoured. The present study was planned to compare the whole body retention and Schilling's test for Vitamin B-12 absorption.

MATERIAL AND METHODS

Subject and Drugs: 12 healthy male volunteers ranging in the age from 20 years to 50 years were included in this study. Peripheral blood examination was carried out to ensure normal hemopoietic function in each subject. Isotope labelled Vitamin B-12 (57Co labelled Vitamin B-12 and 58Co labelled Vitamin B-12) was supplied as capsules by Bhabha Atomic Research Centre, Trombay. Each capsule contained 1 μ g of Vitamin B-12 with specific activity of 1 mci per mg.

Whole body retention studies: One capsule of 57Co labelled vitamin B-12 was given orally to the subject after an overnight fast. The whole body radioactivity was measured with a shadow shield whole body counter with 10.2 cm diameter x 7.6 cm thick sodium Iodide detector. The subject lying on the bed passed under the detector from head to toe in 500 seconds and

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radioactivity was recorded on a spectrometer. For ^{57}Co a single energy band of (75-175) DeV covering the photopeak region was used. The counts were registered in prone and supine position. In pilot studies, the measurements were made immediately and 1,2,3,4,5,6, hours after administration of capsule and then every day for 8 days. Subsequently measurements were made at 4 hours and on 4th day after the administration of capsule. Subjects were not allowed to pass urine during first 4 hours.

Schilling's test :

Dose preparation (2) : 1 μg of Vitamin B-12 (labelled) was dissolved in 22 ml water and out of this, 2 ml was used for preparation of standard 20 ml was given to volunteers on empty stomach after an overnight fast. One mg of non radioactive vitamin B-12 was injected intramuscularly two hours later. Urine was collected for 24 hours and radioactivity was measured in a well-type counter. The percentage of radio activity excreted in urine was calculated from the appropriate standard. The whole body measurements were also performed as above.

Whole body retention test was done first on each volunteer using low energy ^{57}Co isotope and a week later Schilling's test was performed on the same subject using ^{58}Co .

RESULTS

Fig. 1 shows the hourly whole body radioactivity measurements in two representative subjects after giving oral labelled vitamin B-12. It is seen that the activity shows increase in first one or two hours and then remains steady. There is very little change in body radioactivity after 4 hours. Therefore 4 hour body radioactivity was taken as 100 percent for the calculation of absorption of vit. B-12 (7).

Fig. 2 shows the body radio activity on days 1 to 7 in five subjects. Radioactivity at hour after giving labelled Vitamin B-12 is taken as 100 percent. It will be seen that radioactivity goes on decreasing upto the 3rd day, but there is no change after the 4th day indicating that this is the radioactivity due to stored vitamin B-12. Therefore the percentage of radioactivity present on 4th day can be taken as the nearest approximation of absorbed vitamin B-12 (7).

Table I shows the estimates of vitamin B-12 absorption on 12 subjects with Schilling's test and whole body retention. It will be seen that whole body retention test (column D) gives much higher estimates of Vitamin B-12 absorption as compared to Schilling's test (column A). It is also significant that in Schilling's test after giving the flushing dose considerable radioactivity is retained in the body (column B) but it is less than that retained without flushing dose (column D).

The sum of columns A & B (column C) comes close to the figures in column D but are lower in all cases except one. In some cases these are much lower & discrepancies are probably due to errors in 24 hours urinary collection.

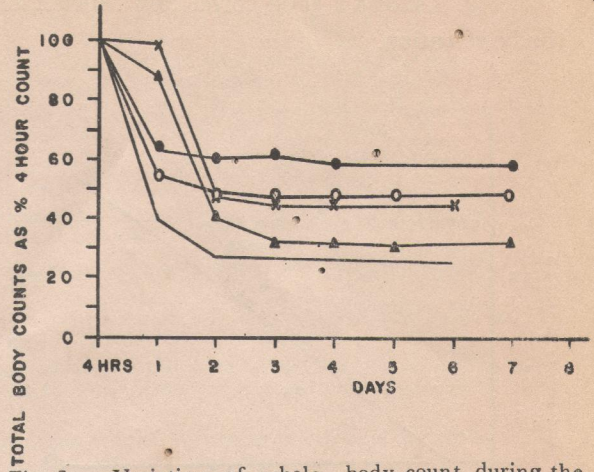
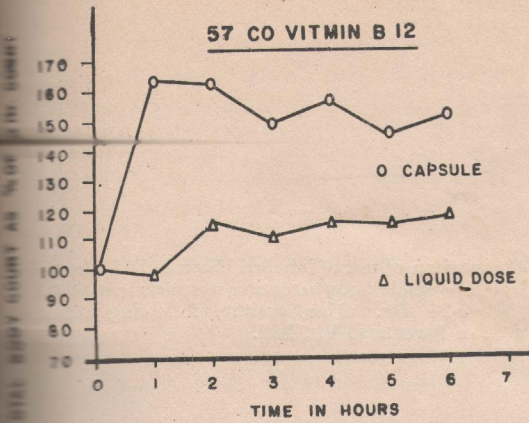


Fig. 1: Shows hourly whole body radioactivity measurements in two subjects after giving oral dose of labelled Vitamin B12.

Fig. 2: Variation of whole body count during the period 4 hrs to 7 days after the administration of oral dose of radioactive Vitamin B12.

Table I: Shows the estimates of vitamin B12 absorption on 12 subjects with Schilling's test and whole body retention.

No. of volunteer	Schilling's Test			Whole body test
	24 hours urinary excretion of 58 Co B12	Whole body retention after flushing dose	Absorption of 58 Co B12 AS (A+B)	Absorption of 57 Co B12 without flushing dose
	A	B	C	D
1	21.7%	38.7%	60.4%	70.5%
2	24.5%	31.2%	55.7%	75.5%
3	23.2%	22.6%	45.8%	78.0%
4	18.8%	15.4%	34.2%	49.0%
5	30.0%	32.7%	62.7%	67.0%
6	8.6%	17.6%	26.2%	30.0%
7	7.8%	20.0%	27.8%	45.0%
8	9.7%	21.2%	30.9%	32.0%
9	17.4%	28.1%	45.0%	54.0%
10	7.8%	22.0%	29.8%	41.3%
11	24.6%	52.2%	76.8%	66.7%
12	9.4%	26.2%	35.6%	69.2%

Fig. 3 shows a good correlation between the urinary excretion in Schilling's test and whole body retention.

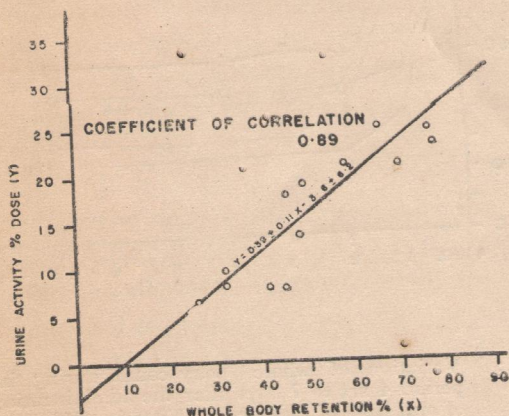


Fig. 3: Comparison of test of labelled Vitamin B12 absorption by (X)=whole body retention measurement at 4 days (Y)=0-24 Hr. urine activity following a flushing dose of Parenteral Vit. B12.

DISCUSSION

The relationship between the 24 hour urinary excretion and 4th day retention of vit. B-12 consistent with these parameters being in simple proportion for an individual. This relationship has also been shown by Cottrel *et al.* (4) and whole body counting provided more accurate data on absorption of vit. B-12.

Though Schilling's test gave lower estimate of absorption, there was good correlation between the two. This was probably because there was a good correlation between the absorbed Vit. B-12 and its excretion in urine.

However, in view of the difficulties in collecting urine for 24 hours accurately, whole body retention appeared to be a method of choice. Also, when the estimates were lower in normal subjects, the discrimination between poor absorbers and normal ones also became less sensitive. Thus on both these counts whole body retention measurements are claimed to be more acceptable in addition to being more accurate.

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