

Vitamin B₁₂ Absorption from Eggs (38940)

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(Introduced by H. H. Zinneman)

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The absorption of radiocyanocobalamin incorporated into meat protein *in vivo* has been studied in normal subjects after ingestion of physiologic doses of vitamin B₁₂ (1). The assimilation of the vitamin equaled or surpassed that observed in controls who ingested comparable amounts of labeled crystalline B₁₂ (2). In contrast, the absorption by normal volunteers of physiologic doses of ⁵⁷Co B₁₂ incorporated into eggs by injection into hens *in vivo* was rather poor when the eggs were in the form of eggnog (3) or scrambled (cooked) (4). The great difference between the assimilation of B₁₂ from meat and from eggs is unknown. One explanation may be the differences in the technique for labeling the B₁₂-containing food (1, 3, 4).

Because the absorption of vitamin B₁₂ from eggnog was slightly superior to that from scrambled eggs a second possibility should also be considered, namely, that the form in which eggs are prepared may influence the absorption of vitamin B₁₂. A study based on this possibility is described in the following report.

Materials and Methods. The subjects of this study were 18 healthy volunteers who had gastric acid present as determined by tubeless gastric analysis in 15 and with gastric intubation after betazole hydrochloride stimulation in 3. All subjects had normal serum vitamin B₁₂ concentrations ranging from 200 to 700 pg/ml and normal absorption of aqueous crystalline ⁵⁷Co B₁₂. They were served comparable portions of ⁵⁷Co B₁₂ eggs cooked in three ways—scrambled, boiled, or fried—and similar portions of aqueous crystalline ⁵⁷Co B₁₂. The method for obtaining the ⁵⁷Co B₁₂ eggs is described in detail elsewhere (4). The amount of radio-B₁₂ absorbed after ingestion of each of the egg mixtures and of the crystalline ⁵⁷Co B₁₂ was measured by stool and urinary excretion tests and 8-hr plasma absorption determina-

tions of radioactivity (5-7) as earlier described (4). These values were then compared.

The scrambled eggs were prepared two ways, with whole eggs and with yolks only, because most of the vitamin B₁₂ content of eggs is in the latter (8, 9). The preparation of both mixtures was identical. The uniformity of the two mixtures was ascertained; the details have been published elsewhere (4).

The scrambled egg mixtures contained 0.4-0.5 μ Ci of ⁵⁷Co and 0.56 μ g of vitamin B₁₂ for the scrambled whole eggs and 0.5 μ g for the yolks. The aqueous crystalline ⁵⁷Co B₁₂ contained 0.56 μ g vitamin B₁₂ and 0.5 μ Ci of ⁵⁷Co. The allotted amount of vitamin B₁₂ was present in 45-56 g of scrambled whole eggs and in 15 g of yolks as determined by *Euglena gracilis* assay (4, 10). The equal portions of labeled eggs were individually packaged in cellophane bags and stored at -20° until served. Reheating in a microwave oven for 30 sec had no deleterious effect on the vitamin B₁₂ content.

The boiled and fried eggs were prepared so that the whites were coagulated and the yolks were still mainly liquid. They were served with a piece of toast and a cup of coffee for breakfast after an overnight fast as were the portions of scrambled whole eggs and yolks. The aqueous crystalline ⁵⁷Co B₁₂ was also served after an overnight fast (4).

The individual whole eggs contained about 0.5 μ Ci of ⁵⁷Co, but their individual vitamin B₁₂ content could not be measured. These eggs weighing about 60 g each were obtained from two different hens that were fed a standard Purina Flock Chow diet and produced eggs with an average vitamin B₁₂ content of 0.67 μ g (range 0.51-0.82) and 0.85 μ g (range 0.72-0.94), determined on six and seven eggs, respectively. The vitamin B₁₂ measurements were performed on eggs

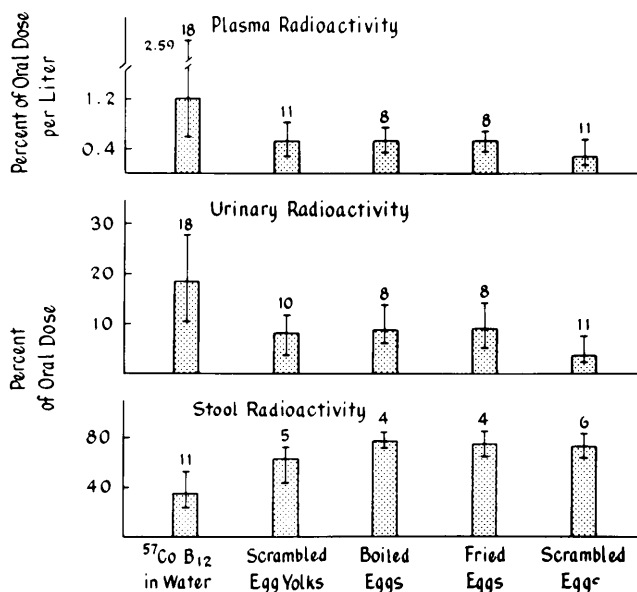


FIG. 1. Comparison of plasma radioactivity and urinary and stool excretion values after ingestion of crystalline ⁵⁷Co B₁₂ dissolved in water or incorporated into eggs *in vivo* and served in different forms to healthy subjects. Number of tests performed are shown above the bars, which indicate the average values with upper and lower ranges.

obtained immediately before the injection of ⁵⁷Co B₁₂, and the absorption tests were given at intervals of a week or more.

When individual eggs were given to the subjects, the radioactivity was measured by a Tobor scintillation counter. This value represented 100% of the ingested ⁵⁷Co in the stool excretion test samples. In the urinary excretion and plasma absorption test results, radioactivity was determined indirectly by Tobor scintillator measurement of 10 ml of water containing 0.56 μg of B₁₂ and 0.5 μCi of ⁵⁷Co. A 1% aliquot of this dose, also dissolved in 10 ml of water, was measured in the same well-type scintillation counter as used for counting plasma and urine. A correction factor was then used to compute the total count of each individual egg ingested. The urinary and stool excretion values were expressed as a percentage of the ingested ⁵⁷Co, and the plasma absorption value was expressed as a percentage of ⁵⁷Co present in a liter of plasma.

Results. The average values with ranges for the three absorption tests can be seen in Fig. 1. The results of the urinary excretion tests showed that the average assimilation of ⁵⁷Co B₁₂ from scrambled egg yolks (8.02%),

boiled eggs (8.91%), and fried eggs (9.19%) was quite similar and slightly more than twice that assimilated from scrambled whole eggs (3.66%) ($P < 0.01$), but slightly less than half that absorbed from aqueous crystalline ⁵⁷Co B₁₂ (18.62%) ($P < 0.1$). The plasma absorption values confirmed those of the urinary excretion tests. The average stool excretion values for the four forms of eggs served (63.6, 76.0, 75.8, 72.5%, respectively) showed rather poor absorption compared with the crystalline ⁵⁷Co B₁₂ (35.2%) ($P < 0.01$). In contrast to the urinary and plasma tests, the stool excretion method showed no statistical difference between the various forms of eggs served ($P > 0.10$). This may be due to the small number of subjects who collected stool, the small difference in absorption, or to the difficulty in obtaining accurate stool collection for a whole week.

Discussion. The results of the urinary and plasma tests show that the form in which the eggs are prepared may influence the absorption of vitamin B₁₂. It remains to be seen whether this observation is unique for eggs. Assuming that the absorption of nonlabeled vitamin B₁₂ in eggs is similar to that of ⁵⁷Co B₁₂, the nutritional vitamin B₁₂ value of

boiled and fried eggs is superior to that of scrambled eggs.

The contents of the three types of cooked eggs were identical, but the average ⁵⁷Co B₁₂ absorption from the scrambled eggs was less as judged by the urinary excretion and the plasma tests; therefore, the degree of absorption must in some way be related to the method of preparation. Two possible explanations may be suggested. One, a better assimilation of vitamin B₁₂ from liquid rather than from coagulated egg yolk is unlikely since the absorption of B₁₂ from boiled and fried eggs was no better than from scrambled (cooked) egg yolks. The second, an inhibitory effect on B₁₂ absorption by ovalbumin, is more likely and supported by the fact that the absorption of ⁵⁷Co B₁₂ from scrambled whole eggs was inferior to that of scrambled egg yolks.

The absorption of vitamin B₁₂ from eggs served in any form was much inferior to that of a comparable amount of crystalline ⁵⁷Co B₁₂. The reason for the difference is unclear, but several possibilities are suggested. Essential for the absorption of vitamin B₁₂ is its attachment to the gastric intrinsic factor (IF), a process that occurs primarily in the stomach (11, 12). Given in free form, aqueous crystalline ⁵⁷Co B₁₂ immediately combines with IF in gastric juice. ⁵⁷Co B₁₂ incorporated into food, however, is bound to protein and thus is not immediately available to combine with IF.

A certain amount of digestion in the stomach seems to be required to release protein-bound vitamin B₁₂ for attachment to IF although few facts regarding this stage of vitamin B₁₂ assimilation are available (12). Even in the best of circumstances, digestion in the stomach may not be sufficiently complete to release all the protein-bound vitamin B₁₂ to IF, and the process may be completed in the intestine. However, the vitamin B₁₂-rich bile may cause isotopic dilution of the ⁵⁷Co B₁₂ and lead to incomplete absorption of the labeled vitamin B₁₂.

The naturally occurring form(s) of vitamin B₁₂ in eggs and the form(s) of the injected ⁵⁷Co B₁₂ are unknown. It has been shown that the two naturally occurring coenzymes of vitamin B₁₂, i.e., methylcobalamin and 5'-

deoxyadenosylcobalamin, are not as readily absorbed as cyanocobalamin and hydroxocobalamin (13-17). If vitamin B₁₂ in eggs is present in coenzyme form(s) and little or no conversion to hydroxocobalamin occurs during the food preparation, this may at least in part explain the lower absorption rate of egg vitamin B₁₂.

Eggs are known to impair the absorption of iron (18-20), and a substance in ovalbumin, avidin, inhibits absorption of biotin (21). The possibility that eggs may contain an inhibitor of B₁₂ assimilation should therefore, also be considered.

Although the absorption of vitamin B₁₂ from scrambled egg yolk and boiled or fried eggs was superior to that from scrambled whole eggs, it was still inferior to that reported to be incorporated into meat *in vivo*. The reason for the great difference between the degree of assimilation of vitamin B₁₂ from meat and from eggs remains to be found.

Summary. The assimilation of ⁵⁷Co B₁₂ from *in vivo* labeled eggs was much inferior to that of a comparable amount of crystalline ⁵⁷Co B₁₂. Furthermore, the absorption varied with the form in which the eggs were served. Judged by the urinary excretion test and the plasma absorption of radioactivity the average absorption from boiled and fried eggs was more than twice that from scrambled whole eggs, but less than half that absorbed from crystalline ⁵⁷Co B₁₂.

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