but otherwise the narcotic effect was judged unaltered.

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Further Observations on Distribution of Radioactivity Following Parenteral Administration of Co^{60} Vitamin B_{12}.* (22529)

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It is now well established that following oral or parenteral administration of Co^{60} vit. B_{12} radioactivity may be detected over the liver, spleen and left kidney for periods up to 315 days(1-4). It has been further demonstrated that following the intramuscular injection of $Co^{60}Cl_2$ the pattern is entirely different, suggesting that uptake studies with the radiovitamin probably represent the B_{12} molecule in the organs surveyed(4).

The present report is concerned with further investigations on the distribution of Co^{60} vit. B_{12} under a variety of conditions: (1) following intravenous injection of the radiovitamin; (2) in association with large doses of "cold" B_{12} before, simultaneous with, or after intramuscular injection of the radiovitamin; and (3) administration of amethopterin prior to intramuscular injection of the radiovitamin.

Material and methods. The Co^{60} vit. B_{12} had a specific activity of 0.750 $\mu c/\mu g$. Each dose was diluted with sterile distilled water so that one ml contained one μg of radiovitamin. All administered doses were one ml in volume. Intramuscular injections were given in the right deltoid muscle, using a 1-inch 22 gauge needle. "Cold" vit. B_{12} was administered in the deltoid muscle of the opposite arm. A well-shielded sodium iodide crystal scintillation counter having an aperture of 2 cm was placed over (1) the site of injection (right deltoid muscle), (2) precordium, (3) lower right chest (liver), (4) lower left chest (spleen), (5) left lumbar region (left kidney), (6) lower end of sacrum, (7) lateral aspect of thigh. Radioactivity was recorded for 5-minute periods over each site within ½, 1, 2, 3, 4, 8, 12 and 24 hours after injection. Thereafter, measurements were made at 2, 3, 5 and 7-day intervals up to 168 days. Background counts were subtracted from observed values. No correction was made for Co^{60} decay. All patients were inmates of a chronic disease hospital and received essentially the same diets. Their ages varied from 43 to 80 years. The diagnoses included hypertensive heart disease (1), bronchial asth-

* Co^{60} vit. B_{12} and "cold" B_{12} were supplied by Merck and Co., Rahway, N. J. Amethopterin was supplied by Lederle Laboratories, American Cyanamid Co., Pearl River, N. Y.

TABLE I. Organ Distribution of Radioactivity following Parenteral Administration of Co^{60} Vit. B_{12}.

Treatment	No. of patients	Disappearance from site of inj. (days)	Days observation	Duration of plateau (days) Liver	Spleen	Kidney
A. Co^{60} B_{12}, intrav.	4	--	161–168	24–111	4–106	1–34
B. Co^{60} B_{12}, I.M. & cold B_{12} inj. for 7 days previously	2	3	120–127	104–113	120–127	6– 7
C. *Idem* & cold B_{12}, same time	4	½–3	53– 84	—	—	—
D. " " after 1 hr	2	1	13– 27	13– 27	6– 12 hr	3–12 hr
E. " " " 2 "	2	3–4	48– 70	7– 68	5– 12 "	5– 6 "
F. " " " 24 "	3	3–5	106	1– 91	3– 54	2 hr–20 days
G. " " " 7 days	4	—	86–101	86–101	64–101	1–87

ma (2), reticulum cell sarcoma (1), chronic lymphocytic leukemia (1), pulmonary fibrosis (1), rheumatoid arthritis (4), arteriosclerosis (8), gout (2), cirrhosis of liver (1), luetic heart disease (1), unilateral exophthalmus (1), and familial telangiectasia (1).

Results. The results are tabulated in Table I. Since no significant levels of radioactivity were noted over the precordium, sacrum and thighs, these sites were excluded from the table. The first column under each organ records the time after injection of the radiovitamin when maximal number of counts was noted.

(1) *Intravenous Co^{60} vit. B_{12}.* Four subjects received 1 μg of Co^{60} vit. B_{12} intravenously (Table I-A). Maximal activity appeared over the liver, left kidney and spleen in about 30 minutes, in contradistinction to hours and days respectively, noted following muscular injection(4). In all other respects the plateau and duration of activity in all 3 organs were similar to those noted following intramuscular administration.

(2) *Intramuscular Co^{60} vit. B_{12} associated with "cold" B_{12}.* (a) Two patients received 30 μg of "cold" vit. B_{12} for 7 days. This was followed by the intramuscular injection of 1 μg of Co^{60} vit. B_{12} on the eighth day (Table I-B). The data show that the pattern of distribution of radioactivity over liver, spleen and kidney was essentially the same as that previously reported from this laboratory when no "cold" B_{12} was administered. (b) Four subjects received 1 μg of Co^{60} vit. B_{12} intramuscularly in the right deltoid muscle. Immediately thereafter 1 mg of "cold" B_{12} was injected into the left deltoid muscle (Table I-C). The pattern of distribution of radioactivity was entirely different from those previously observed(4). Radioactivity disappeared from the site of injection in 12 to 72 hours. It reached its maximum over the liver in 2 to 3 hours. This level was of a low order, and only slightly above background counts. This low level of activity over the liver persisted for the duration of the study in each subject (up to 84 days). Radioactivity reached a maximum over the spleen in a short time (2 to 3 hours) but was again only slightly above background counts. Except for one case all detectable activity was gone in 4 days and no significant plateau was attained. Similar observations were made over the left kidney; *viz.*, maximal appearance in 2 to 3 hours, no plateau, and rapid disappearance over the left kidney in 3 subjects (6 hours to 2 days) and prolonged activity of 55 days in one case. (c). Two subjects received 1 μg of Co^{60} vit. B_{12} in the right deltoid muscle. This was followed 1 hour later by the intramuscular injection of 1 mg of "cold" B_{12} in the left deltoid muscle (Table I-D). The observed results were similar to those noted in experiment 2(b). Disappearance of radioactivity from the right deltoid muscle was delayed for 24 hours. The height of maximal activity over liver, spleen and left kidney never rose above the 1 hour post-injection level of the radiovitamin. The plateau was lower than that observed in previously described experiments(4). The plateau over the liver was maintained in both subjects for the duration of the experiment. Over the spleen and left kidney the plateaus were all short (maximum 12 hours), but ac-

tivity was detectable during the periods of observations. (d). Two patients received 1 μg of Co^{60} vit. B_{12} in the right deltoid muscle followed by 1 mg of "cold" B_{12} in the left deltoid muscle 2 hours later (Table I-E). The results in this study resemble those observed in experiment 2(c). Disappearance of the radiovitamin from the right deltoid muscle was delayed up to 96 hours after injection. Maximal activity over the liver was attained after 2 days, which was at a lower level than that generally observed in previous experiments(4), and only slightly higher than in experiment 2(c). Although radioactivity was detectable over the liver for the entire period of observation (48 and 72 days respectively) the duration of the plateau was extremely short in 1 case (7 days). (e). Three subjects received 1 μg of Co^{60} vit. B_{12} in the right deltoid muscle followed by 1 mg of "cold" B_{12} in the left deltoid muscle 24 hours later (Table I-F). Disappearance of radioactivity from the deltoid muscle was apparently not affected by the large dose of "cold" B_{12} as it fell within the range previously reported from this laboratory(4). However, maximal activity over the liver never rose above the 24 hour level and the plateaus in 2 of the 3 cases were only 1 and 3 days, respectively. In the third patient maximal activity was attained 1 day after injection of "cold" B_{12} in 1 patient, 2 hours later in another, and never rose above the preinjection level in the third. The plateau of radioactivity varied from 3 to 54 days. Detectable counts were present during the entire period of the experiment. Over the left kidney no further rise in counts was observed after administration of "cold" B_{12}. The plateau of radioactivity varied from 2 hours to 20 days. However, activity was present from 90 to 106 days. (f). Four subjects received 1 μg of Co^{60} vit. B_{12} in the right deltoid muscle. Seven days later 1 mg of "cold" B_{12} was injected into the right deltoid muscle. (Table I-G). The pattern of distribution and plateaus and duration of activity over liver and spleen were the same as those previously reported from this laboratory when no "cold" B_{12} was administered(4). Over the left kidney the plateau ranged from 1 to 86 days. Such extreme patterns were seen only in experiment 2(b), when 1 mg of "cold" B_{12} was given simultaneously with 1 μg of Co^{60} vit. B_{12}.

(3) *Prior administration of amethopterin.* Three subjects received 10 mg of amethopterin for 7 days, following which 1 μg of Co^{60} vit. B_{12} was injected into the right deltoid muscle. The patterns of disappearance of activity from the site of injection, uptake, plateau and duration over the liver, spleen and left kidney were similar to those previously reported from this laboratory when no folic acid antagonist was administered(4).

Discussion. From the foregoing results it appears that following intravenous injection of 1 μg of Co^{60} B_{12} the pattern of distribution of radioactivity over the liver, spleen and left kidney is similar to that observed after intramuscular injection. As might be anticipated, the time of initial and maximal appearance is earlier, probably because of direct introduction of the radiovitamin into the circulation.

The experiment in which the subjects received 30 μg of "cold" B_{12} for 1 week prior to intramuscular injection of Co^{60} B_{12} was set up with the purpose of "saturating" the organs with the vitamin, anticipating a slow or poor uptake of the radiovitamin. However, these results were not observed, suggesting that in the 24 hours between the last injection of "cold" B_{12} and the intramuscular injection of Co^{60} B_{12} enough vitamin was released from the liver, spleen and kidney to permit maximal deposition from 1 μg of Co^{60} B_{12} in the organs named. No corroboration for this hypothesis is, however, available. The studies with injection of Co^{60} B_{12} simultaneously with, or preceding, the administration of 1 mg of "cold" B_{12} by 1, 2 and 24 hours show what was expected; namely, a dilution effect of the large dose of non-radioactive vitamin on the absorption from site of injection and its deposition in liver, spleen and left kidney. Some of the inconsistencies in duration of plateau noted in the earlier report from this laboratory(4) were again observed, particularly in persons receiving the "cold" B_{12} 24 hours after the radiovitamin. The unanswered problem is the failure of the thousandfold

dose of "cold" B_{12} to dilute the radiovitamin so much so that none of it would be deposited or retained in the organs described. In the group of patients who received 1 mg of "cold" B_{12} 7 days after the injection of the radiovitamin there was no alteration in the pattern or plateau over the liver, spleen or kidney during the period of observation (up to 86 days). This would suggest that vit. B_{12}, once it reaches these organs, is not readily displaced by further administration of the vitamin.

The experiment in which amethopterin was administered for one week prior to Co^{60} B_{12} was investigated because of the intimate relationship of folic acid and vit. B_{12} in nucleoprotein synthesis. However, no alteration in the pattern of radioactivity distribution was observed. It is possible that a longer period of folic acid antagonist administration is necessary to affect vit. B_{12} deposition.

Conclusions. (1) Prior administration of 30 μg of "cold" B_{12} or 10 mg of amethopterin daily for 1 week, or injection of 1 mg of "cold" B_{12} 7 days after intramuscular injection of 1 μg of Co^{60} vit. B_{12}, does not alter rate of disappearance from site of injection or pattern of distribution of radioactivity over liver, spleen and left kidney. (2) Intravenous injection of 1 μg of Co^{60} B_{12} is followed by a more rapid appearance of radioactivity over the organs named. (3) Intramuscular injection of 1 mg of "cold" B_{12} simultaneous with 1, 2 or 24 hours after the administration of 1 μg of Co^{60} B_{12} reduces height of radioactivity over organs named but does not prevent its localization there.

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Role of Various Tissues in Metabolism of C^{14} Labeled Priscoline in the Rat.* (22530)

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Priscoline (tolazoline) hydrochloride is well known today, clinically as an effective peripheral vasodilating agent and pharmacologically as an adrenergic blocking agent. In a previous study, C^{14} labeled Priscoline administered to rats was rapidly excreted in the urine almost entirely unchanged(1). That a small amount of the drug is changed in the body was suggested by recovery of traces of C^{14} in the expired air and by the separation of small amounts of a radioactive metabolite in the urine. The method of analysis described in the present study permitted the detection of minute amounts of radioactive Priscoline and this metabolite in tissues as well as in urine, and made possible further investigation of the roles of various tissues in the turnover of Priscoline.

Methods and materials. The Priscoline used in this study had a specific activity of 1 μc/mg, and was labeled at the single benzyl carbon between the phenyl and imidazoline rings.‡ Sprague-Dawley albino rats weighing from 167 to 253 g were injected intraven-

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