Correlation of Serum Binders to Clearance Rates of Intravenously Administered ^{57}Co B_{12}[1] (35353)

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^{57}Co B_{12} administered intravenously to some pernicious anemia patients in relapse and to chronic myelocytic leukemia subjects was cleared from plasma at a slower rate than when administered to normal subjects (1, 2). It is known that in chronic myelocytic leukemia and some cases of pernicious anemia serum transcobalamin I content is elevated (3–5). The present study was undertaken for the purpose of correlating percentage residual activity of intravenously injected ^{57}Co B_{12} to transcobalamin I and II content in serum.

Materials and Methods. Plasma clearance. 0.1 μCi of ^{57}Co B_{12} in 1 ml was injected intravenously into 10 subjects. Blood samples were drawn at 1, 2, 5, 15, 30, and 45 min and 1, 2, 4, 8, 12, 24, and 48 hr. Aliquots of 4 ml of plasma were counted in a scintillation counter and percentage residual activity was calculated from estimated plasma volume and radioactivity of the injected sample (2).

DEAE-cellulose column chromatography. One-ml serum aliquots from samples obtained during clearance studies were chromatographed on 5.0-ml DEAE-cellulose columns as described by Retief *et al.* (5). The method was modified in that eleven 2.0-ml samples were collected with the 0.06 *M* phosphate buffer (pH 6.35), and six 2.0-ml samples with 1.0 *M* NaCl. DEAE-cellulose (purchased from Schleicher and Schuell, Keene, N.H.) of ion exchange capacity between 0.90 and 0.95 meq/g was used.

Regression equation. The abbreviated Doolittle method was employed in deriving

TABLE I. Measured and Estimated Percentage Residual Activity in Plasma 1 hr After the Injection of ^{57}Co B_{12}.

Subject	X_1[a]	X_2[b]	X_3[c]	Residual activity (%) Measured	Residual activity (%) Estimated
CAR	20	80	128	15	14.2
HEB	16	84	126	15	13.2
ING	21	79	110	14	13.4
MAT	22	78	130	12	14.8
MEL	18	82	65	8	9.9
SAE	21	79	79	13	11.4
POP	25	75	59	11	11.0
KIV	26	74	117	11.4	13.8
FIG	15	85	160	11.8	15.0
SHA	15	85	160	13.7	15.0

[a] X_1 = % of added ^{57}Co B_{12} bound to transcobalamin I when 100 pg of labeled vitamin were added to 1 ml of serum.

[b] $X_2 = (100 - X_1)$.

[c] X_3 = pg of ^{57}Co B_{12} bound to transcobalamin I when 2000 pg of ^{57}Co B_{12} were added to 1 ml of serum.

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the multiple regression equation (6). Correlation coefficient was calculated from the formula $R = (\Sigma\hat{y}^2/\Sigma y^2)^{1/2}$.

Results and Discussion. The following equation was derived from the data shown in Table I:

$$\hat{y} = 11.3 + 0.12X_1 - 0.09X_2 + 0.06X_3$$

where $\hat{y}$ = estimated residual radioactivity in plasma at 1 hr; X_1 = percentage of added ^{57}Co B_{12} bound to transcobalamin I when small amounts (100 pg/ml) of vitamin were added to serum; X_2 = percentage of ^{57}Co B_{12} bound to transcobalamin II determined under the same conditions as X_1; X_3 = pg of ^{57}Co B_{12} bound to transcobalamin I when 2000 pg of ^{57}Co B_{12} were added to 1 ml of serum. The multiple correlation coefficient for the set of data shown in Table I was 0.74.

It appears that the residual activity in plasma of intravenously injected ^{57}Co B_{12} is correlated to the distribution of the labelled vitamin among the binders and the amount of transcobalamin I present in serum.

Summary. An equation was derived which permits the calculation of the residual radioactivity in plasma at the end of 1 hr following intravenous injection of ^{57}Co B_{12}. This equation gives the residual activity on the basis of the distribution of *in vitro* added ^{57}Co B_{12} to serum vitamin B_{12} binders as determined by DEAE-cellulose column chromatography.

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