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Relative Effectiveness of B₁₂ and Dimethylbenzimidazole B₁₂-Coenzyme in Thymine Methyl Biosynthesis.* (25863)

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We have recently reported that Vit. B₁₂ stimulates conversion of formate to 5-methyl group of thymine by chick bone marrow cells (1). Further studies have indicated that the vitamin functions in reduction of one-carbon compounds between formate and formaldehyde levels of oxidation(2,3). Barker *et al.*(4) reported isolation of coenzyme forms of B₁₂ which catalyze conversion of glutamate to β -methylaspartate by certain microorganisms. In this system B₁₂ is inactive. It appeared desirable to determine effectiveness of the coenzyme form of B₁₂ in stimulating conversion of formate to thymine methyl.

Methods. Handling of chicks, preparation of bone marrow cell suspensions, details of incubation and formation of the acetol osazone from thymine were the same as previously described(1). Bone marrow cells from B₁₂-deficient chicks were incubated with formate-C¹⁴ and deoxyuridine and various concentrations of Vit. B₁₂ or 5,6 dimethyl-benzimidazole B₁₂-coenzyme(4). The results are reported as per cent increase in conversion of formate-C¹⁴ to thymine as a result of addition of B₁₂ or the coenzyme.

Results. Each result given in Table I is the average of at least 2 experiments. It is quite clear that the coenzyme form of B₁₂ was less effective than the crystalline vitamin in stimulating conversion of formate to the 5 methyl group of thymine. This observation

TABLE I. Influence of Vit. B₁₂ and 5, 6 Dimethylbenzimidazole B₁₂ Coenzyme (DMBC) on Conversion of Formate C¹⁴ to Thymine Methyl by B₁₂-deficient Chick Bone Marrow Cells. (Results expressed as % increase in conversion of formate to thymine methyl.)

mg/ml	B ₁₂	DMBC
15	35	9
150	66	34
1500	90	43
3750		47

is in agreement with other reports indicating that the coenzyme form of B₁₂ does not stimulate methionine biosynthesis under conditions in which the vitamin is effective(5,6). It appears that Vit. B₁₂ has at least 2 distinct metabolic roles, one in reduction of one carbon compounds and the other in biochemical rearrangements catalyzed by coenzymes first described by Barker *et al.*(4).

Summary. Vit. B₁₂ was more effective in stimulating conversion of formate to thymine methyl by chick bone marrow cells than was 5,6 dimethylbenzimidazole B₁₂-coenzyme.

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