Cobyric Acid from Vitamin B_{12}

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COBYRIC ACID (cobyrinic acid *abcdeg* hexa-amide, factor V_{1a}) is of interest as the goal in the total synthesis of vitamin B_{12}, and as the starting material for the partial chemical synthesis of certain modifications of the vitamin which possess high antivitamin activity.[1] An important source of this compound has been sewage sludge: we have now found that under certain hydrolytic conditions cobyric acid may be obtained directly from vitamin B_{12}.

The hydrolysis of vitamin B_{12} with dilute acid or base does not generally yield detectable amounts of cobyric acid, presumably because the propionamides *b*, *d*, and *e* are hydrolysed preferentially.[2] However, under carefully controlled conditions of high acidity (*e.g.* 11·3 N–HCl, 65°, 18·5 min.) hydrolysis of the primary amide groups is somewhat suppressed and after appropriate purification steps (DEAE–cellulose column chromatography, followed by preparative paper chromatography of the monocarboxylic acids) cobyric acid is isolable in ~10% yield. The identification of the product rests on comparison with an authentic sample in the following respects:—ultraviolet and visible spectrum, infrared spectrum, paper chromatography (six systems), paper electrophoresis (pH 2, 7, and 10); and on the conversion of the compound into cobinamide using the mixed anhydride route.[3] By-products from the hydrolysis include cobinamide and other neutral compound, probably factor Ia,[4] and a dicarboxylic acid fraction. The 5,6-dimethyl-1-α-D-ribofuranosylbenziminazole 2'- and 3'-phosphates are associated with the latter, and may be readily separated from it.[5] Hence both cobyric acid and the mixed nucleotides required for partial synthesis are available from the one reaction.

It seems likely that the procedure described depends on two assisted cleavages. The 2'-hydroxyl group of the ribose assists in the cleavage of the amino-alcohol–phosphate linkage giving cobinamide and (initially) the 2',3'-cyclic phosphate. Accordingly cobinamide is obtained in good yield using a shorter reaction time.[3] Secondly the β-hydroxyl group of the aminopropanol participates in the cleavage of amide *f* (giving cobyric acid and Dg-1-amino-2-propanol with retained configuration[6]) possibly *via* rearrangement to the isomeric ester. The preferential cleavage of seryl and threonyl residues in peptides subjected to highly acidic conditions is an analogous reaction.

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