The Temperature Dependence of some Physical Properties of Cobinamides and Cobalamins

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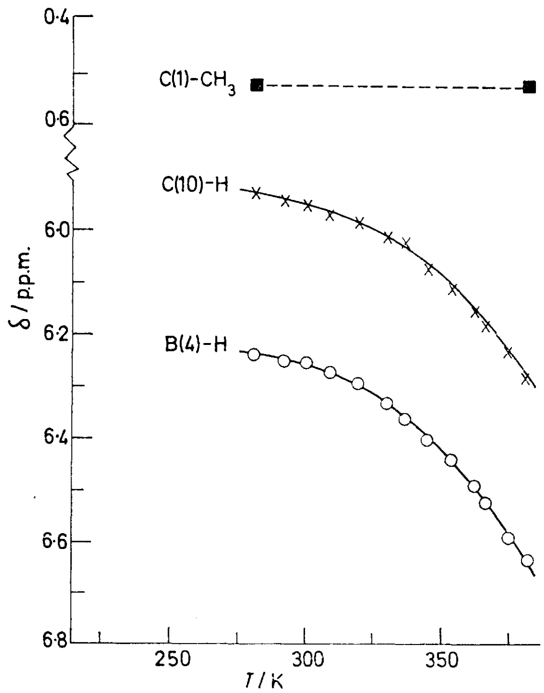
The temperature dependences of the electronic absorption spectra and the ^1H n.m.r. spectra of the cobinamides and cobalamins are discussed. A conformational change, which does not involve the removal of the 5,6-dimethylbenzimidazole ligand from the co-ordination sphere of the cobalt is favoured as an explanation of the temperature dependence of these physical properties of the cobalamins.

In previous papers[1-4] we discussed the temperature dependence of a number of spectroscopic properties[5,6] of the cobinamides and cobalamins. With certain cobinamides it was apparent that there existed an equilibrium between two species, called[2] the ' low- ' and ' high-temperature ' forms. This information derived principally from marked changes in the electronic absorption spectra and ^1H n.m.r. spectra with temperature, the change being reflected in a change of colour from ' red ' at low temperature to ' yellow ' at high temperature. The extent to which this equilibrium lies to the ' red ' or ' yellow ' side depends markedly on the axial ligand, the ratio of the ' red ' to ' yellow ' forms increasing in the order $CN^- < CH_2{=}CH^- < CH_3^- < C_2H_5^-$. It was suggested[1-4] that the equilibrium involved a six-co-ordinate (low-temperature, red) form and a five-co-ordinate (high-temperature, yellow) form. It was considered[7] that an unidentified conformational change would accompany the change in co-ordination number. As stated[4] previously, the description of a



The temperature dependence of the chemical shifts of the C(10)–H, B(4)–H, and C(1)–CH_3 of coenzyme B_{12} in ^2H_2O

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[2] R. A. Firth, H. A. O. Hill, B. E. Mann, J. M. Pratt, R. G. Thorp, and R. J. P. Williams, *J. Chem. Soc.* (*A*), 1968, 2419.

[3] H. A. O. Hill, J. M. Pratt, and R. J. P. Williams, *Discuss. Faraday Soc.*, 1969, **47**, 165.

[4] H. A. O. Hill, J. M. Pratt, and R. J. P. Williams, *Chem. in Britain*, 1969, **5**, 156.

[5] J. M. Pratt, 'Inorganic Chemistry of Vitamin B_{12},' Academic Press, London, 1972.

[6] H. A. O. Hill, ' Corrinoids, in Inorganic Biochemistry,' ed. G. L. Eichhorn, Elsevier, Amsterdam, 1973, p. 1067.

[7] See especially the discussion in ref. 3 and ref. 4.

complex as five-co-ordinate must include a complex in which the sixth ligand is at a longer equilibrium distance from the metal as compared with the six-co-ordinate species. Obviously this can happen only if accompanied by a change in structure of the rest of the complex.

In the discussion of the temperature dependence of the electronic spectra of the cobalamins, it was suggested [2] that, by analogy with the cobinamides, the high-temperature form of coenzyme B_{12}, 5′-deoxyadenosylcobalamin, was a species in which the 5,6-dimethylbenzimidazole was not co-ordinated to the cobalt. An analysis of the temperature dependence (see the Figure) of the chemical shifts of the C(10)–H and the B(4)–H indicates that 25% of the complex is in the high-temperature form. We have now completed [8] an assignment of *ca.* 70% of the ^{1}H n.m.r. spectrum of coenzyme B_{12}. In particular we have assigned the C(1)–CH_3 to the resonance at 0.53 p.p.m. It is shifted up-field by the ring current of the 5,6-dimethylbenzimidazole. Consequently it is the resonance most affected by the removal of the 5,6-dimethylbenzimidazole from the co-ordination sphere of the cobalt by protonation or, *e.g.* in methylcobalamin, by cyanide. The chemical shift of this resonance is *not* temperature dependent. Hence it is difficult to reconcile this observation with the suggestion that, in the high-temperature form, the 5,6-dimethylbenzimidazole is not co-ordinated to the cobalt. Therefore we conclude that, in the case of the cobalamins, the temperature-dependence results from a conformational change. We agree with Hogenkamp *et al.*[9] that this conformational change involves a change in the ‘ pucker ’ of the corrin ring.

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[8] O. D. Hensens, H. A. O. Hill, J. Thornton, A. M. Turner, and R. J. P. Williams, *Proc. Roy. Soc.*, in the press.

[9] H. P. C. Hogenkamp, P. J. Vergamini, and N. A. Matwiyoff, *J.C.S. Dalton*, 1975, preceding paper.