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Possible Biochemical Implications of the Crystal Structure of Biotin

Abstract. An examination of the molecular architecture of biotin, as determined by x-ray crystallographic analysis, has indicated that biotin may be capable of forming an intramolecular hydrogen bond in solution. A review of various chemical analogs of the vitamin has shown a close correlation between the possibility of forming such a hydrogen bond and biotin-like activity.

A recent x-ray analysis of the crystal structure of biotin (*1*) has established the stereochemistry of the molecule and, in particular, shown it to have the *cis-cis* configuration at the three asymmetric carbon atoms (Fig. 1). Indications that both the stereoisomerism (*2*) and the length of the aliphatic chain (*3, 4*) are specific for biological activity prompted a detailed examination of the molecular structure, in the hope that this might throw some light on the mode of action of the vitamin.

An accurate scale model of the biotin structure and—for comparison—analogous models incorporating the alternative configurations at the asymmetric centers and aliphatic chains of several different lengths were constructed. Though the ring portions of the models were rigid, flexibility was allowed in the construction of the side chains so that the effects of rotation about carbon-carbon single bonds might be examined.

While in general the various interatomic distances and angles of the biotin molecule conform with those found in similar structures, there are some unusual features near the junction of the ring and chain portions of the molecule. There is a particularly short separation (2.8 A) between atoms C_{10} and N_7, and a C_8–C_9–C_{10} angle of 119°. This unusually large angle, which is presumably the result of repulsion between C_{10} and N_7, would appear to facilitate rotation in solution about the C_9–C_{10} single bond, which would otherwise be restricted by steric hindrance. When the ring and chain portions of the biotin model were folded together, by a rotation about the C_9–C_{10} bond, it was found that the chemically reactive centers in the ureido ring system and the carboxyl group could approach each other closely, while Van der Waal's distances of separation were maintained between the other atoms of the chain and the ring system. In particular it was found that such a folding, together with only small rotations about other single bonds in the chain, would enable the structure to meet the rather stringent physical requirements for hydrogen bonding between O_6 and one of the carboxyl oxygen atoms (Fig. 2) (*5*).

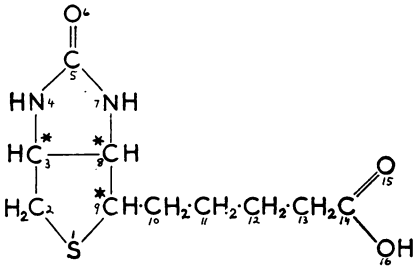

Fig. 1. Structural formula of biotin (atoms numbered arbitrarily).

A study of the various other models indicated that the short C_{10}–N_7 separation in biotin (and presumably the large C_8–C_9–C_{10} angle) is a direct consequence of the *cis-cis* configuration. Furthermore, none of the three stereoisomers of biotin, or molecules with different chain lengths, appear to be capable of forming an intramolecular hydrogen bond, the possibility of which depends critically on both the steric configuration and the chain length.

Supporting evidence for the implication of this type of hydrogen bonding in the biological function of the vitamin appears to be provided by studies of the biotin-like properties of several dozen chemical analogs of the vitamin. These studies have indicated a high degree of biological specificity for the structure of biotin, not only with regard to the steric configuration (*2*) and the length of the aliphatic chain (*3, 4*), but also with regard to the ureido ring system (*6*) and the presence of an oxygen atom at the position of the carboxyl group (*4*). However, it is possible to modify the ring containing the sulfur atom (*7*) and to prepare amides and amino acid derivatives of biotin (*8*)—neither of which need necessarily prevent intramolecular hydrogen bonding—without destroying the biological activity.

It is not quite clear how the formation of an intramolecular hydrogen bond would affect the chemical reactivity of the molecule. In aqueous solution such a hydrogen bond would presumably be unstable, allowing the biotin molecule to alternate between two different states. The formation of the hydrogen bond might be expected to alter the charge distribution in the ureido ring system and to displace the keto-enol equilibrium to enol, resulting in a change of chemical reactivity at the nitrogen atoms, or a system of hydrogen transport along the lines suggested by Lichstein (*9*), whereby the substrate may donate a proton at one point and accept one at another during a keto-enol transition (*10*).

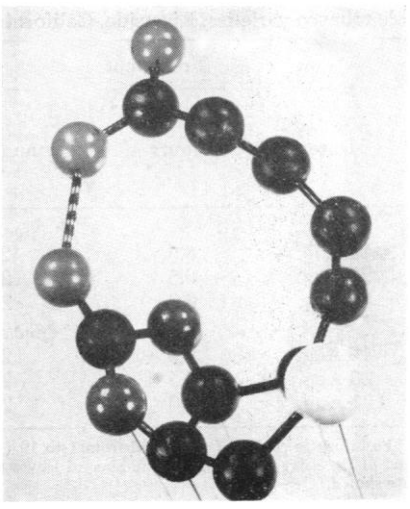

Fig. 2. Possible mode of intramolecular hydrogen bonding in biotin: O_{16} lies in the plane of the ureido ring system; O_6 lies in the plane of the carboxyl group; the distance O_{16}–O_6 is about 2.6 A, and all the other distances between atoms of the chain and those of the ring system (except N_7–C_{10}) are greater than 3.4 A. Angles C_{14}–O_{16}–O_6 and C_5–O_6–O_{16} are both about 120°.

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