PHOTOSENSITIZED MONOMERIZATION OF 1,3-DIMETHYLURACIL DIMERS

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The formation of pyrimidine dimers in DNA and RNA has been reported to account for part of the chemical changes caused by ultraviolet radiation in biological systems.[1,2] Cleavage of thymine dimers to the monomer moieties is regarded as the chemical reaction involved in the photoreactivation process performed by enzymes in the presence of visible light.[3] The enzyme-sensitized photoreactivation process, which leads to the repair of the major part of the photochemical lesion in DNA, has received much attention; however, its detailed mechanism is still obscure. A possible mechanism for this process is a photosensitized rupture of the pyrimidine dimers through an energy transfer process (a), or through formation of a complex between the enzyme and the substrate (b).

(a) $S \xrightarrow{h\nu} S^*$

$S^* + \widehat{MM} \longrightarrow S + [\widehat{MM}]^*$

$[\widehat{MM}]^* \longrightarrow 2M$

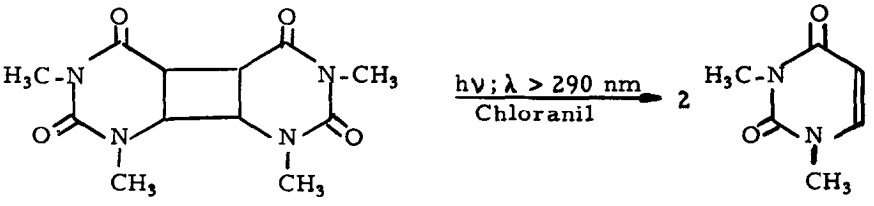
(b) $S + \widehat{MM} \longrightarrow S \text{-----} \widehat{MM} \xrightarrow{h\nu} [S \text{ -- } \widehat{MM}]^*$

$[S \text{ -- } \widehat{MM}]^* \longrightarrow S + 2M$

S = Photosensitizer; M = Monomer; $\widehat{MM}$ = Dimer

The photomonomerization of pyrimidine dimers by the use of chemical photosensitizers could serve as a most useful model for studying the mechanism involved in the enzyme-sensitized photoreactivation and the chemical reactions involved in this process.[4] In addition, the application of photosensitized-rupture to a group of isomeric dimers may lead to the possible selective cleavage of a certain type of dimer, and to the development of selective monomerization agents specific to the appropriate dimers.

We used the four dimers of 1,3-dimethyluracil (DMU) as models for pyrimidine dimers and chloranil as the chemical photosensitizer. The four dimers of DMU were found to be stable toward ultraviolet irradiation of $\lambda > 290$ nm (Pyrex filter) and could be monomerized directly only by short ultraviolet light ($\lambda \sim 240$ nm).[5] We have found that irradiation of the DMU dimers in the presence of chloranil employing ultraviolet light of $\lambda > 290$ nm led to different rates of cleavage of three dimers, B, C and D, while the fourth dimer, the trans-anti, dimer A, remained practically intact under these conditions of irradiation.



three isomers

The three dimers were converted to monomer to different extents under similar reaction conditions. Thus, dimer B was monomerized to the extent of 10%, C to 8%, and D to 50%.

The four DMU dimers were prepared by irradiation of DMU in t-butanol-acetone mixture with light of $\lambda > 290$ nm.[5] The photosensitized monomerization of the dimers was effected by irradiation at room temperature of a solution of the dimer (500 mg.) in a 1:1 chloroform-methylenechloride mixture (150 ml.) in the presence of chloranil (500 mg.) for 20 hr. Oxygen was excluded from the reaction mixture by a stream of nitrogen bubbling through the solution. The progress of the reaction was followed by examining the absorption at 265 nm. Isolation of the products was achieved by column chromatography on silica gel in petroleum ether-acetone mixtures, and the monomers isolated were identified by direct comparison with authentic samples (m.p., mixed m.p., R_f in TLC, IR and NMR spectra). No DMU could be detected in mixtures of its dimers and chloranil when kept in the dark for the same length of time. Neither could any monomer be detected when DMU dimers in chloroform-methylenechloride

solutions in the absence of chloranil were irradiated with light of $\lambda > 290$ nm (Pyrex filter).

Ben-Hur and Ben-Ishai have recently found[6] that the yeast photoreactivating enzyme in the presence of visible light cleaved the cis-syn but not the trans-syn thymine dimer in irradiated denatured DNA. Our results indicate that the employment of a chemical photosensitizer also leads to a selective photorupture of isomeric pyrimidne dimers, and might serve as a key for cleavage of only certain pyrimidine dimers in a polymeric chain.

It is noteworthy that quinones have been connected with photoprotection and repair of damage in irradiated DNA. Our observations might shed light on the role of the quinones in this process.[7]

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