UV-Induced Charge Separation in DNA and Sequence Selective Reactions

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UV-light induces different photochemical reactions in DNA which may lead to mutations or cell death. After several 10^9 years of evolution today's living organisms possess sophisticated repair systems, which allow them to deal with photo-lesions. In earlier stages of evolution however, simpler repair mechanisms may have played a role and may have influenced the development of the genetic code. In this study, we report on the discovery of the simplest possible, sequence selective repair mechanism[1] which acts without any need of external structures, e. g. proteins, cofactors or DNAzymes.[2]

Photoexcitation of nucleobases in single and double stranded DNA is known to produce very short-lived excited singlet states as well as charge-transfer states living on the tens to hundreds of picoseconds time scale. These relatively long-lived reactive states are generally thought to induce photodamages.[3,4] However we could recently show that such states can also repair the cyclobutane pyrimidine dimer (CPD) photo-lesion by electron transfer in a photolyase-like reaction. The molecular mechanism resembles strongly the process used by photolyase, an important class of DNA repair enzymes. In analogy to the photolyase, photoexcitation of DNA in the vicinity of a cyclobutane pyrimide dimer (CPD) lesion initiates the repair by a reactive charge transfer state. These states are one of the major decay channels of photoexcited DNA.

In our experiments we used time resolved UV-pump, IR-probe spectroscopy to characterize the light-induced charge-transfer states of different dinucloetides. We use UV-illumination of a variety of short single and double stranded oligonucleotides with and without a T=T CPD-lesion and monitor photo-destruction or photo-repair by UV- spectroscopy and HPLC. We could show that photoexcitation of a single adenine (A) adjacent to the T=T-dimer does not lead to CPD-repair. However, excitation of a guanine (G) adenine (A) sequence reverts the T=T CPD-lesion to the intact TT bases. The involvement of two bases in the repair indicates that the long-living charge transfer state G^+A^- between G and A is responsible for the repair process. The charge-transfer process has been investigated for a number of dinucleotides where the respective cation and anion could be detected via infrared bands. For GA (see Fig. 1) one observes a long-lived (ca. 300 ps) radical-pair state G^+A^-.[5] Shorter lifetimes for radical-pair states are observed for other dinucleotides, e.g. 50 ps for T^-A^+.[6,7] From G^+A^- the A radical anion donates an electron to the CPD, which induces splitting of the cyclobutane ring and reformation of the intact T bases.[1] A number of different sequences

adjacent to the T=T CPD lesion are investigated, supporting the idea of repair via electron transfer from the transiently formed charge transfer states. The experiments show that the mere presence of an adjacent radical pair is not sufficient for CPD repair. More likely the redox potential of the radical ion pairs and the life times of the charge separated states are the driving power to control the repair. Repair capacities are found to be strongly sequence dependent, creating regions on a DNA strand with different tendencies of self-repair or self-destruction. Such processes may have played an important role in the evolutionary selection of UV-exposed DNA sequences in the prebiotic world where other repair mechanisms were missing.

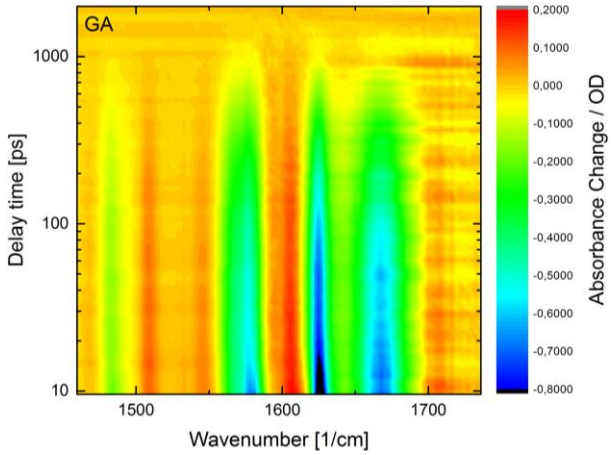

Figure 1. Time-resolved IR absorption changes after 266 nm excitation of GA. 3D plot of the experimental data corrected for long-lasting absorbance changes.

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