CHEMICAL AND STRUCTURAL PROPERTIES OF DNA-ABASIC SITE CROSS-LINKS

Nathan Price

Dr. Kent Gates, Dissertation Supervisor

ABSTRACT

The loss of a coding nucleobase from the structure of DNA is a common event that generates lesion known as an abasic (Ap) site. Ap sites exist as an equilibrating mixture of a cyclic hemiacetal and a ring-opened aldehyde. Aldehydes are electrophilic functional groups that can form covalent adducts with nucleophilic sites in DNA. Thus, Ap sites present a potentially reactive aldehyde as part of the internal structure of DNA. Here we report evidence that the aldehyde group of Ap sites in duplex DNA can form a covalent adduct with the N^6-amino group of adenine residues on the opposing strand. The resulting interstrand DNA-DNA cross-link occurs at 5'-ApT/5'-AA sequences in remarkably high yields (15-70%) under physiologically relevant conditions.

In order to rigorously characterize the structure of dA-Ap cross-links by enzymatic digestion of cross-linked DNA a putative dinucleoside cross-link remnant was synthesized by reaction of dA with 2-deoxyribose. LC-MS/MS analysis provided evidence that the synthetic compound 6 has the same chemical structure as the cross-link remnant released by enzymatic digestion of cross-linked DNA. Additionally, this work demonstrates that cross-links arising from the reaction of an Ap-site with an opposing nucleobase occur in a multitude of sequence context. Understanding the structure and reactivity of Ap-derived cross-links may be very important as this naturally-occurring DNA-templated reaction has the potential to generate cross-links in the genetic material of living cells.