Poster Presentation

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Structural basis for error-free bypass of bulky adduct by DNA polymerase Polκ

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Humans are frequently exposed to the environmentally ubiquitous and potentially carcinigenic polycyclic aromatic hydrocarbon, benzo[a]pyrene (BP). BP is metabolized to highly reactive benzo[a]pyrene diol epoxides (BPDEs) in the cells. BPDEs react with DNA predominantly at the N2 position of guanine and form bulky adducts. The major BP adduct is (+)-trans-anti-[BP]-N2-dG (BP-N2-dG) that is carcinogenic. The bulky adduct block DNA synthesis by replicative or high-fidelity DNA polymerases. Some of the specialized lesion bypass polymerases (mostly belonging to Y-family) can replicate through this bulky adduct but often in an error prone manner, resulting in mutagenesis. Among the four human Y-family polymerases Polη, Polκ, Polι and Rev1, Polκ is unique in its ability for efficient and error-free replication through BP induced BP-N2-dG adduct. In this study, we determined the crystal structures of human Polκ (hPolκ) in ternary complex with DNA and an incoming nucleotide dCTP analogue. The crystals contain DNA with either G base or (+)-trans-anti-[BP]-N2-dG adduct at a template-primer junction and diffract to 2.5 Å and 2.8 Å, respectively. The structures reveal that hPolκ is able to accommodate the bulky adducted DNA in its minor groove without base flipping and nucleotide looping out. The bulky adduct has the polycyclic BP moiety in the minor groove in the regular helical conformation. Polκ has a unique active site that is more open at the minor groove side than other Y-family polymerases. The damaged guanine is in the anti-conformation, the dCMPNPP incoming nucleotide maintains normal Watson-Crick pairing with the G* base. This is the first structure of eukaryotic Y-family polymerase carrying the minor groove BP adduct. The structure and biochemical analysis provides a basis for understanding how hPolκ can correctly bypass and tolerate BP induced BP-N2-dG adduct in human cells.

Keywords: Y-family DNA polymerase, DNA polymerase kappa, benzopyrene