208 DNA repair function and structural modeling of a putative DNA glycosylase NEIL3

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Mammalian genes, *NEIL1*, *NEIL2*, and *NEIL3*, have been discovered as *E. coli* Nei/Fpg glycosylase family. Although repair activities of NEIL1 and NEIL2 for oxidative damage have been characterized, enzymatic activity of NEIL3 remains unclear. Here, we present a structural model for NEIL3 and examine the recombinant protein on its possible DNA repair activity. We find no clear glycosylase activity of NEIL3 as well as its DNA glycosylase domain to several types of DNA substrate containing a modified base. Nevertheless, NEIL3 retains DNA binding activity and AP-lyase activity. Moreover, NEIL3 can partially rescue *E. coli nth nei* mutant from hydrogen peroxide sensitivity. These results suggest that NEIL3 can work as a DNA glycosylase to repair detrimental base lesions generated by oxidative stress *in vivo*.

209 Molecular analysis of mitochondrial DNA in solid organs of irradiated rats

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Our laboratory previously reported a concordant increase of relative mitochondrial DNA (mtDNA) content and the number of large-scale mtDNA deletions in radiation-associated post-Chernobyl papillary thyroid carcinoma. In this study, we investigated the same indices in several kinds of tissues of irradiated rats, to determine whether mtDNA might be used as a bioindicator of a genotoxic exposure. Five-week old rats were acutely irradiated with single doses of 0.5 and 1 Gy of X-rays, and sacrificed at the age of 6 months. All solid organs show that specific number of large-scale deletions in mtDNA does not change significantly in any tissue, in irradiated or control animals, as normalized for the relative mtDNA content. Our data suggest that the number of large-scale deletions in mtDNA and mtDNA levels in a given tissue does not appear to be associated with exposure.

210 Mutation Spectrum of *Bacillus subtilis* Spore Irradiated With Heavy Ion Near the Bragg Peak

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High LET radiation such as heavy ions, are predicted to induce densely clustered DNA lesions unlike low LET radiations. We are interested in the possibility of the difference of the mutational spectra due to the LET difference. *Bacillus subtilis* strain HA101 spores were irradiated with Ar and Fe ions near the Bragg peak. From irradiated spores, 41 mutants exhibiting rifampicin resistance were isolated, and together with 25 spontaneous mutants, sequence changes in the *rpoB* gene were determined. Among the spontaneous ones, 24 single-base substitutions (SBS) and 1 three-base insertion were found. Among the irradiated ones, 37 SBS, 2 tandem-double substitutions, 1 three-base insertion and 1 double substitution skipping 6 bases were found. Among 37 SBS, 20 were an identical substitution from G:C to A:T forming a hot spot. So far, no difference in base substitution spectra between Ar and Fe ions was detected. Further investigation with low LET radiations are in progress.

211 Effect of RibCys on scavenging long-lived radicals and reducing gene mutation in mammalian cells by adding AFTER irradiation

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We report that RibCys (2(R,S)-D-ribo-1′,2′,3′,4′-tetrahydroxybutly-thiazolidene-4(R)-cariboxylic acid), previously shown to protect against radiation induced pathology in rats and pigs, scavenges long-lived radicals (LLR), mainly in proteins assigned as sulfinyl radicals (R-CH_2SO). LLR levels were directly measured by electron spin resonance (ESR) spectroscopy at 77 K in Syrian Golden Hamster embryo (SHE) cells un-irradiated or gamma-ray irradiated (5 kGy) at room temperature. RibCys (480 mM) was added 2 h after irradiation. The rate constant between LLRs and RibCys was estimated as $3.2 \times 10^{-4}\,M^{-1}s^{-1}$, about 20 times slower than for vitamin C. In mutation experiments, RibCys (4 mM) added after radiation with carbon ions (3 Gy) decreased levels of $CD59^-$ mutants in Chinese hamster ovary hybrid A_L cells by about 50%, compared with 70% with Vitamin C. These results provide further evidence that LLR are mutagenic and may trigger genomic instability.