

P048 Analysis of DNA repair in living cells

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DNA lesions arising from environmental and endogenous sources induce various cellular responses including cell cycle arrest, DNA repair and apoptosis. Although detailed insights into the biochemical mechanisms and composition of DNA repair pathways have been obtained from *in vitro* experiments, a better understanding of the interplay and regulation of these pathways requires DNA repair studies in living cells. We employed laser microirradiation and photobleaching techniques in combination with specific mutants and inhibitors to analyze the real-time accumulation of proteins at laser-induced DNA damage sites *in vivo*, thus unravelling the mechanisms underlying the coordination of DNA repair in living cells. In summary, we found immediate and transient binding of repair factors involved in DNA damage detection and signalling, while repair factors involved in the later steps of DNA repair, like damage processing, DNA ligation and restoration of epigenetic information, showed a slow and persistent accumulation at DNA damage sites. We conclude that DNA repair is not mediated by binding of a preassembled repair machinery, but rather coordinated by the sequential recruitment of specific repair factors to DNA damage sites.