DYNAMICS OF HETEROCHROMATIN PROTEIN 1 (HP1) and X-RAY REPAIR CROSS-COMPLEMENTING PROTEIN 1 (XRCC1) IN DNA REPAIR FOCI, REVEALED BY FLIP AND FCS

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Keywords: XRCC1, HP1, DNA damage, FLIP, FCS, dSTORM.

BACKGROUND. Induction of DNA lesions results in recruitment of hundreds of copies of repair factors and formation of DNA repair foci [1-3]. Some repair foci exhibit internal organization [3,4]. XRCC1 is a repair factor recruited to single strand DNA breaks. It has no known enzymatic activity and acts a molecular platform recruiting other repair proteins, including DNA ligase III, polymerase PARP1 and PARP2, and polymerase β. HP1 is recruited to DNA lesions of various types. It is known as a dominant suppressor in position-effect variegation (PEV) effect. Its various post-translationally modified forms are involved in a number of nuclear processes including gene silencing, chromatin remodeling, replication and repair of DNA, but the role of HP1 in DNA repair is not known.
Goal: This study aimed at providing information about dynamics of XRCC1 and HP1 in repair foci in order to formulate hypotheses regarding potential roles played by the high numbers of these proteins at the sites of DNA damage (SSB and DSB).
Methods: Cells expressing mRFP-XRCC1 (including a protein with mutated domains BRCT1 and BRCT2) or eGFP-HP1 were studied, local DNA damage was induced by focused visible light without the use of photosensitisers [5]. Protein dynamics was measured using FLIP and FCS.
Results: In the nucleoplasm only two subpopulations of HP1 and XRCC1 exist, one diffusing freely, another transiently bound to chromatin. Within the repair foci three subpopulations of HP1 can be detected in DNA, one being highly dynamic, and two characterized by low mobilities. Two subpopulations of XRCC1 can be detected in repair foci. Both are much less dynamic than XRCC1 in the nucleoplasm outside of repair foci.
Conclusions. Large numbers of copies of HP1 and XRCC1 recruited to DNA single- and double-strand breaks are bound in the vicinity of DNA lesions and their mobility is restricted. The hypothesis which assumes that HP1 and XRCC1 may stabilize chromatin in the vicinity of DNA lesions will be discussed.

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