PARP1 retention by PARP inhibitors delays downstream repair events and drives cytotoxicity

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Poly(ADP)-ribose polymerase 1 (PARP1) and PARP2 are the major sensors of single-strand DNA breaks (SSBs), involved in multiple repair pathways as well as replication fork protection. PARP inhibitors (PARPi) are employed in the treatment of ovarian, breast, pancreatic, and prostate cancer, capitalizing on the principle of synthetic lethality with deficits in homologous recombination (HR), most notably BRCA1 and BRCA2 deficiency. The cytotoxicity of PARPi has been largely attributed to their capacity for catalytic inhibition of PARP1/2 and their ability to prolong the association of PARP1 with DNA breaks, effectively “trapping” the enzyme on damaged chromatin. This trapping effect manifests as a decreased turnover of PARP1 molecules at the site of damage. In principle, PARPi are believed to induce the accumulation of highly toxic double-strand DNA breaks that cannot be repaired in the absence of effective HR, thus triggering cell death. While the PARPi mechanism of action has been extensively interrogated, the contribution of PARP1 catalytic inhibition and trapping to cytotoxicity and the relationship between these two factors remain elusive. We introduced three parameters for the characterization of PARPi-induced changes in PARP1 dynamics at sites of micro-IR-induced complex DNA damage. These include the PARP1 Inhibition Coefficient (PIC), the PARP1 Trapping Coefficient (PTC), and the PARP1 Retention Coefficient (PRC), which are assigned to an inhibitor of interest. Herein, we demonstrate that the PRC correlates with the delayed recruitment of multiple representative factors implicated in multiple pathways that converge for the repair of complex DNA lesions. In particular, inhibitor-induced PARP1 retention at breaks decouples PCNA removal from RPA recruitment, suggestive of a dysregulated sequence of repair events and compromised proficiency. Finally, we show that the capacity for PARP1 retention is largely associated with the cytotoxicity of evaluated PARPi *in vitro*.