Enzyme-free colorimetric assay of uracil DNA glycosylases activity using toehold-mediated DNA strand displacement event

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Herein, we describe a novel enzyme-free strategy for colorimetric uracil DNA glycosylases (UDG) activity assay, which relies on toehold-mediated DNA strand displacement circuit. The strategy utilize dsDNA, which undergo denaturation in the presence of UDG and release catalyst strand that triggers sequential toehold-mediated strand displacement reaction. This event leads to the release of a split G-quadruplex DNAzyme strand, which is initially blocked and inactivated by a blocker strand. Furthermore, a fuel strand is employed for the recycling of the catalyst strand to promote another toehold-mediated strand displacement event, consequently producing a large number of active split G-quadruplex DNAzymes. By employing this strategy, we sensitively determined the UDG activity based on the distinct colorimetric signal promoted by the ABTS oxidation developed by the peroxidase mimicking activity of the released G-quadruplex DNAzymes.