Fluorescent G-quadruplex probe for the assay of base excision repair enzyme activity

이창열, 박기수, 박현규†
한국과학기술원
(hgpark@kaist.ac.kr†)

Herein, we describe a novel strategy to accurately determine the uracil DNA glycosylase (UDG) activity by utilizing G-quadruplex probe incorporating 2-aminopurine (2-AP), a fluorescent nucleobase analog. The assay relies on hybridized duplex DNA consisting of a 2-AP incorporated signaling strand and a uracil incorporated strand, from which UDG removes the uracil bases and dissociates the duplex DNA into single-stranded DNAs. This phenomenon enables the formation of the G-quadruplex structure of the signaling strand, consequently leading to significant fluorescence enhancement of 2-AP within the G-quadruplex. Based on the enhanced fluorescence, we have reliably determined the activity of UDG and also demonstrated the capability of this strategy to screen candidate UDG inhibitors. The proposed design could have a great potential to serve as a universal platform in the development of novel systems to assay many other enzymes involved in the base excision repair pathway.