CRISPR-Cas12a-assisted cell-free DNA biosensor based on Metal-Enhanced Fluorescence and Colorimetric Analysis

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Cell-free DNA (cfDNA) can be used as a biomarker for cell death and defects due to its low concentration in circulating blood. However, application of cfDNA in clinical fields is not easy because of difficulty in nucleic acid amplification step. In this study, nucleic acid amplification free-CRISPR-Cas12a-assisted cfDNA biosensor was fabricated using DNA-associated gold nanoparticle (GNP) for a metal-enhanced fluorescence (MEF). The fabricated cfDNA biosensor was designed to cleave single-stranded DNA (ssDNA) between FIIC and larger GNP when presence of a target cfDNA (BRCA-1). The dissociation of the larger GNP from FIIC changed color from purple to red-purple. As a result, this cfDNA biosensor can detect BRCA-1 presenting high sensitivity (as low as 0.34 fM) and rapid (<30 min) detection. The developed method can be adapted to quantify levels of other nucleic acids as a promising diagnostic and prognostic strategy in a field-deployable platform. Acknowledgments: This research was supported by the National Research Foundation of Korea (NRF) grant funded by the Korea government (MSII) (No. 2019R1A2C3002300) and by the Ministry of Education (No. 2016R1A6A1A03012845).