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Hybrid CRISPR/Cas protein for one-pot detection of DNA and RNA

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Abstract

Clustered regularly interspaced short palindromic repeats (CRISPR)-based diagnostics have emerged as next-generation molecular diagnostics. In CRISPR-based diagnostics, Cas12 and Cas13 proteins have been widely employed to detect DNA and RNA, respectively. Herein, we developed a novel hybrid Cas protein capable of detecting universal nucleic acids (DNA and RNA). The CRISPR/hybrid Cas system simultaneously recognizes both DNA and RNA, enabling the dual detection of pathogenic viruses in a single tube. Using wild-type (WT) and N501Y mutant severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) as detection models, we successfully detected both virus strains with a detection limit of 10 viral copies per reaction without cross-reactivity. Furthermore, it is demonstrated the detection of WT SARS-CoV-2 and N501Y mutant variants in clinical samples by using the CRISPR/hybrid Cas system. The hybrid Cas protein is expected to be utilized in a molecular diagnostic method for infectious diseases, tissue and liquid biopsies, and other nucleic acid biomarkers.

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