Short Commentary

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CRISPR/Cas - Prospective Rapid Diagnostic Technique for COVID-19

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The COVID-19 pandemic, triggered by SARS-CoV-2 virus, has severely impacted the living and working conditions of billions of people worldwide. The urgent need for accurate and rapid diagnosis of SARSCoV-2 infection remains critical as global healthcare systems continue to operate during the course of this pandemic.

Currently, the COVID-19 tests kits which are commercially available fall into two major categories. Category one includes detection of SARS-CoV-2 viral RNA using molecular assays such as polymerase chain reaction (PCR) or nucleic acid hybridization-related techniques [1]. Category two involves serological and immunological assays that detect antibodies or antigenic proteins in infected individuals [2]. Development of rapid, cost- effective, point-of-contact test kits providing accurate with timely diagnosis, effective treatment of infected individuals and containing the spread of the virus are the keystones of management.

Presently, there is no definite treatment or vaccination for prevention of COVID-19. Infected patients in critical conditions receive symptomatic or supportive care and clinically stable individuals are isolated [3]. Diagnosis is made based primarily on the symptoms of the disease and qRT-PCR (Quantitative reverse transcription polymerase chain reaction) technique, approved by CDC and World Health Organization (WHO), the COVID-19

(SARS-CoV-2) gold standard detection method [4,5]. The main drawbacks of this technique are the long turnaround time of approximately 4 hours and quantitative analysis requiring bulky instrumentation.

Hence, researchers have recently proposed a coronavirus rapid detection method based on CRISPR/Cas system [6]. CRISPR-Based Assays - Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR) represents a family of nucleic acid sequences found in prokaryotic organisms, such as bacteria. These sequences can be recognized and cut by a set of bacterial enzymes, called CRISPR-associated enzymes, exemplified by Cas9, Cas12, and Cas13. Certain enzymes in the Cas12 and Cas13 families can be programmed to target and cut viral RNA sequences [7]. Two companies, Sherlock Biosciences and Mammoth Biosciences, established by the CRISPR pioneer scientists, are independently exploring the possibility of using the gene editing CRISPR methodology for detection of SARS-CoV-2.

The Specific High-Sensitivity Enzymatic Reporter Unlocking (SHERLOCK) method uses Cas13 that is capable of excising reporter RNA sequences in response to activation by SARS- CoV-2-specific guide RNA. The targets are the S and ORF1ab protein genes in coronavirus genome [8]. The DNA Endonuclease-Targeted CRISPR Trans Reporter (DETECTR) assay by Mammoth Biosciences relies on the cleavage of reporter RNA by Cas12a to specifically detect SARS-CoV-2 viral RNA sequences of the E and N genes, followed by isothermal

amplification of the target, resulting in a visual readout with a fluorophore [9].

Some key advantages of CRISPR-based methods over qRT–PCR are that, results can be read using paper strips to detect the presence of the SARS-CoV-2 virus without loss of sensitivity or specificity and removing the requirement of complex instrumentation. These tests are both low-cost, have rapid turnaround time and can be performed in as little as one hour. These tests have great potential for point-of-care diagnosis [10-12].

Consequently, CRISPR-based technology could swiftly replace qRT-PCR technique considering the high demand for rapid diagnosis tests in current global pandemic state of COVID-19.

Conflict of Interest

Author has no conflict of Interest.

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