



Using EdvoKit #335 to Simulate Reverse Transcriptase Polymerase Chain Reaction (RT-PCR) Testing for COVID-19 Infection

The virus responsible for COVID-19 infection, SARS-CoV-2, is a single-stranded RNA virus. This means that the genome of the virus is RNA, not DNA, and therefore the virus cannot be detected by traditional PCR, since PCR requires a DNA template. In order to detect the virus using PCR, Reverse Transcriptase (RT) is used to synthesize complementary DNA (cDNA) copies of the RNA genome. A small amount of the cDNA is mixed with Taq polymerase, dNTPs and primers for amplification by PCR. Because RT-PCR is extremely sensitive and can detect very low levels of the virus, it is an ideal assay to detect active COVID-19 infections.

For information on the recommended CDC testing protocol, please visit the CDC website:
<https://www.cdc.gov/coronavirus/2019-ncov/lab/testing-laboratories.html>

Directions to adapt EdvoKit 335 to simulate testing for SARS-CoV-2:

1. Create identities of six patients who may have COVID-19. Factors to include are symptoms, recent travel, and current outbreak locations. As this information is subject to change, we suggest you refer to the CDC website for the most up-to-date information.
2. This kit contains enough reagents to run six RT-PCR reactions. Decide how many positive samples and how many negative samples you would like to have.
 - a. For positive samples, aliquot 25 μ L prepared RNA template into a labeled microcentrifuge tube.
 - b. For negative samples, aliquot 25 μ L RNase-free water into a labeled microcentrifuge tube.
 - c. Be sure to make a note of which patient samples are positive and which are negative.
3. Follow the experimental protocol as outlined on pages 10-13 of the EdvoKit 335 literature.
4. Have your students analyze the patient profiles along with the biological data. Which patients have been infected by Sars-CoV-2? Your students should outline their reasoning in their laboratory report.

Please note that this is a simulation for educational purposes and should not be used as a substitute for testing using the FDA-validated test by a healthcare professional.

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