Resource Summary Report

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COVID-19 SCAN molecular workflow

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Proper Citation

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Protocol Information

URL: https://dx.doi.org/10.17504/protocols.io.bebkjakw

Authors: Lea Starita

Group: Coronavirus Method Development Community

Summary: Molecular Detection of SARS-CoV-2 for SCAN the Greater Seattle Coronavirus Assessment Network .justify:after { content: ""; display:inline-block; width: 100%; } .justify:after { content: ""; display:inline-block; width: 100%; } The SCAN real-time RT-PCR assay for SARS-CoV-2 contains primer/probe sets that target the Orf1b and S genes, designed against the SARS-CoV-2 genome (GenBank: MN908947.3). The primer/probe sets were analyzed in silico for specificity for this specific betacoronavirus by Thermo Fisher. All assays are performed on total nucleic acids extracted from upper respiratory tract swabs, including mid-turbinate and nasopharyngeal (NP) swabs stored in Universal Transport Media. The assay limit of detection of 5.6 (Orf1b) and 12.9 (S) molecules per reaction was determined from serial dilutions of synthetic DNA of the target sequences. These probes reproducibly detected SARS-CoV-2 RNA from cultured virus from USA-WA-1/2020 (BEI resources/ATCC) and from 21 specimens confirmed to be positive by the Washington State Department of health. Specificity was determined by lack of detection of SARS-CoV-2 from specimens that are known to be positive for seasonal coronavirus strains HKU1/NL63 or 229E/OC43 and/or other respiratory viruses. These specimens are further inferred to be negative by the fact that they were obtained in the greater Seattle area prior to the first known reports of SARS-CoV-2 in China. .justify:after { content: ""; display:inline-block; width: 100%; } .justify:after { content: ""; display:inline-block; width: 100%; }

Affiliations: University of Washington

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