REFERENCE NUMBER TO 43-00016

SARSeq: MULTIPLEXED, PARALLEL DETECTION OF SARS-CoV2 VARIANTS AND OTHER VIRUSES IN HIGH-TROUGHPUT

Keywords: SARS-CoV2, COVID-19, variant detection, respiratory viruses, multiplexed PCR, NGS, high throughput, mass & routine testing

INVENTION NOVELTY

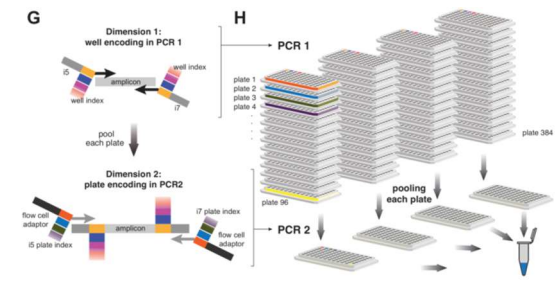
Within a few months, COVID-19, caused by SARS-CoV2, has turned into a global pandemic with severe social and economic consequences. Given the lack of antiviral drugs, vaccination, social distancing, and testing are the only measures to mitigate and track the disease. In an ideal world, mass and routine testing would be the best way to identify active cases as early as possible. Moreover, system critical institutions as well as vulnerable groups of people must be monitored frequently. However, current tests cannot meet the demand for highly sensitive, specific, and accessible surveillance. Molecular diagnostic (MDx) tests represent the "*gold standard*", but are time-consuming and costly, and suffer from global shortage of both equipment and reagents. *SARSeq* provides a highly robust and sensitive test format that facilitates mass and routine testing at low costs and with high accuracy.

VALUE PROPOSITION

SARSeq represents a ground-breaking new concept for the robust, highly multiplexed NGS-based high-throughput detection (e.g., 96x96- or 96x384-well-plate format) of nucleic acids. The method has been validated for the detection of SARS-CoV2 and SARS-CoV2 variants, to provide an efficient and affordable tool for the high-throughput testing e.g., in hospitals, schools, and public services. The method can be also used (either in parallel or separately!) for the detection of any nucleic acid (e.g., derived from viral, bacterial and/or fungal pathogens), and easily adapted to newly emerging pandemics or cancer diagnostics.

TECHNOLOGY DESCRIPTION

SARSeq builds on known methods (i.e., RT, PCR, NGS), but adds tricks, controls, and optimized conditions resulting in a robust and highly accurate MDx test. Test specimens (e.g., gargle or swab) are individually collected and treated with common extraction buffer and heat to release and stabilize RNA. All subsequent steps are conducted in e.g., 96-well-plates. RT generates cDNA using a primer pool of unspecific hexamers and gene-specific 12-mers. During PCR1, amplicons within each well are labeled with two unique well-barcodes, which limits index-hopping and false positives, and end-point PCR is performed to achieve sensitivity across a high dynamic range of virus titers (10^7). All samples from one plate are then pooled, transferred into one well of another e.g., 96-well-plate and PCR2 is conducted wherein specific plate-barcodes along with the adapter sequences for NGS are added. By combining e.g., 96 dual well-barcodes and 96 plate-barcodes, *SARSeq* can be used to generate amplicons from >9,000 (or >36,000 when using 96x384-well plates) samples simultaneously, which are then sequenced in a single, short NGS run.



NGS-based detection pipeline. *Unique dual well-barcodes in PCR1, and specific plate-barcodes in PCR2, allow for simultaneous analysis of amplicons derived from thousands of samples in a single, rapid NGS run (from Yelagandula et al., 2021).*

The primer sets have been optimized to amplify two SARS-CoV2-specific gene fragments and one control. The number of amplicons, however, can be increased to include for instance more virus-specific fragments, controls, or even specific fragments of other pathogens.

COMMERCIAL OPPORTUNITY

SARSeq is available for in-licensing and/or co-development.

DEVELOPMENT STATUS

All aspects of *SARSeq* have been broadly tested, optimized, and compared with conventional systems like SYBR-green-based and Taqman qPCR, as well as other multiplexed NGS tests, validating the method`s robustness and reliability. *SARSeq* received ÖQASTA certificate for the detection of SARS-CoV2 from the Austrian Association for Quality Assurance & Standardization and has been used in Austria since 01/2021 for sequencing the vast majority of SARS-CoV-2 samples.

PATENT SITUATION

Patent applications are currently pending in the US, China, and Europe.

FURTHER READING

Yelagandula et al. (2021) Multiplexed detection of SARS-CoV-2 and other respiratory infection in high throughput by SARSeq. Nat. Commun. 12: 3132-3248.

IMBA Institute of Molecular Biotechnology of the Austrian Academy of Sciences

Ascenion GmbH
Herzogstraße 64
D-80803 München
info@ascenion.de
www.ascenion.de

Licensing Contact
Dr. Torsten Stachelhaus
Technology Manager
T: +49 40 18884348
stachelhaus@ascenion.de

Last updated Oct. 2023