VAMAS

Synthetic Biomaterials
Technical Work Area 40

Call for Participation

Covid-19 Response - 1

Cross-validation of an integrated, phage-derived platform as an open source reference standard for the diagnosis of SARS-CoV-2

Objectives

Performance analysis of an MS2 phage-derived virus-like particle (VLP) platform incorporating the SARS-CoV-2 N gene as an integrated reference material for the diagnosis of SARS-CoV-2. The analysis will provide comparability datasets for:

- MS2-SARS-CoV-2 N gene VLPs by reverse transcription-quantitative polymerase chain reaction (RT-qPCR)
- absolute concentration of the VLP material by droplet digital PCR (ddPCR)

Background

Reference materials are an essential component of nucleic acid-based environmental surveys and medical diagnostic assays. The materials are necessary as process and detection controls as well as reference standards that support the development of diagnostic tools including new primer sets and probes for RT-qPCR, primer sets for loop-mediated isothermal amplification (LAMP), and primer sets and gRNAs for CRISPR-Cas detection.

A number of positive controls are commercially available for the detection of SARS-CoV-2 including synthetic RNA, armoured RNA and the native inactivated virus. However, validated traceable reference materials for the extraction and detection of viral RNA are lacking. MS2 bacteriophage capsids containing packaged N-gene RNA have been developed as a candidate reference material at the London Biofoundry. The performance of the material as an internal standard has been assessed and pre-validated for the detection of SARS-CoV-2 in patient samples in hospital settings.

Standardization needs

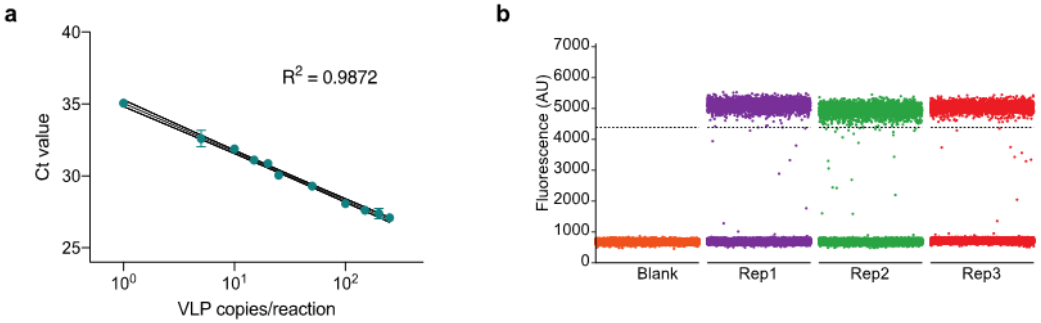
There is an urgent need for an open-source reference material to support the diagnosis of COVID19.

Pre-standardisation needs focus on:

- an integrated VLP reference material for the genetic detection of SARS-CoV-2
- protocols for primary quantitative measurements and data analysis by RT-qPCR using defined primer-probe sets
- protocols for secondary measurements and data analysis for absolute quantification by ddPCR
- performance validation of RT-qPCR measurement values for the material

Work Programme

- MS2-SARS-CoV-2 N gene VLPs together with the protocols of analysis will be distributed to the partners.
- The London Biofoundry will generate, purify, QC and distribute the material in a formulation pre-validated for use in the clinic.



MS2-SARS-CoV-2 N gene VLP reference material. (a) Standard curve generated from a series of RT-qPCR measurements of dilutions of the material. (b) Absolute quantification of MS2-SARS-CoV-2 N gene VLPs in three replicates using ddPCR.

- Aliquots of the material will be distributed to participating laboratories for primary measurement of N gene RNA quantity via RT-qPCR by all participants.
- A secondary measurement of absolute N gene RNA content via ddPCR will be conducted by a select set of participants.
- All results will be compiled and analysed using statistical methods, with full uncertainty evaluation.

Relevant standards & guidelines

ISO20295:2019
ISO 22119:2011
EU2017/746
(EU) 2020/739

Knowledge Transfer

Good practice guidelines, peer-review publications and presentations, and good practice guidelines.

Upon certification, the reference material will be made available through Open MTA to interested parties.

Funding

Participants will fund their own involvement in the project.

Status

The project is active from July 2020. Expression of interest from global participants is welcome.

For more information on participation, please contact:

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