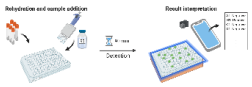
Sep 08, 2020

XPRIZE SHINE - In-tube Fluorescent SARS-CoV-2 Saliva Test



DOI
dx.doi.org/10.17504/protocols.io.bk3rkym6

Jon Arizti-Sanz[1,2], Catherine A. Freije[1], Chloe K. Boehm[1], Sameed M. Siddiqui[1,3], Allen M. Goodman[1], Tinna-Solveig F. Kosoko-Thoroddsen[1], A'Doriann Y. Bradley[1], Jeremy Johnson[1], Pardis C. Sabeti[1,4,5,6,7], Cameron Myhrvold[1,8,7]

[1]Broad Institute of Massachusetts Institute of Technology (MIT) and Harvard, Cambridge, MA 02142, USA.;
[2]Harvard-MIT Program in Health Sciences and Technology, 77 Massachusetts Ave., Cambridge, MA 02139, USA.;
[3]Computational and Systems Biology PhD Program, MIT, Cambridge, MA 02139, USA.;
[4]Harvard T.H. Chan School of Public Health, 677 Huntington Avenue, Boston, MA 02115, USA.;
[5]Department of Organismic and Evolutionary Biology, Harvard University, 26 Oxford Street, Cambridge, MA 02138, USA.;
[6]Howard Hughes Medical Institute, Chevy Chase, MD 20815, USA.;
[7]Massachusetts Consortium on Pathogen Readiness, Boston, MA, USA.;
[8]Department of Organismic and Evolutionary Biology, Harvard University, 26 Oxford Street, Cambridge, MA 02138, USA

XPRIZE Rapid Covid Tes...

Catherine Freije

DOI: https://dx.doi.org/10.17504/protocols.io.bk3rkym6

Protocol Citation: Jon Arizti-Sanz, Catherine A. Freije, Chloe K. Boehm, Sameed M. Siddiqui, Allen M. Goodman, Tinna-Solveig F. Kosoko-Thoroddsen, A'Doriann Y. Bradley, Jeremy Johnson, Pardis C. Sabeti, Cameron Myhrvold 2020. XPRIZE SHINE - In-tube Fluorescent SARS-CoV-2 Saliva Test. **protocols.io** https://dx.doi.org/10.17504/protocols.io.bk3rkym6

Protocol status: Working
We use this protocol and it's working

Created: September 08, 2020

Last Modified: September 08, 2020

Protocol Integer ID: 41809

Keywords: CRISPR, SARS-CoV-2, nucleic acid diagnostic, tube fluorescent assay, tube fluorescent sar, detection of sar, assay, fluorescent, integrated sample inactivation, nasopharyngeal sample, saliva, rna, collected nasopharyngeal sample,

Disclaimer

Abstract

This protocol describes how to perform a SHINE in-tube fluorescent assay to detect SARS-CoV-2 RNA from a self-collected nasopharyngeal sample. This protocol is intended for point-of-care use. All enzymatic components are provided as a single-test freeze-dried pellet for shelf-stable storage, and all steps of the protocol are performed at ambient temperature. The protocol requires a transilluminator or another equivalent blue light emitting device. The protocol presented here is an improved version of the method presented in Arizti-Sanz J*, Freije CA*, *et al.* Integrated sample inactivation, amplification, and Cas13-based detection of SARS-CoV-2. *bioRxiv* (2020).

Image Attribution

biorender.com

Materials

MATERIALS

- Screw cap tube 5 mL sterile **Thomas Scientific Catalog #**1188R46
- FastAmp® Viral and Cell Solution for Covid-19 Testing Solution B **Catalog #**4633
- Reagent Mix A (In-tube SARS-CoV-2 resuspension mix)
- Lyophilized Reagent Mix B (In-tube SARS-CoV-2 detection mix)

STEP MATERIALS

- FastAmp® Viral and Cell Solution for Covid-19 Testing Solution B **Catalog #**4633
- Screw cap tube 5 mL sterile **Thomas Scientific Catalog #**1188R46
- Reagent Mix A (In-tube SARS-CoV-2 resuspension mix)
- Lyophilized Reagent Mix B (In-tube SARS-CoV-2 detection mix)

The necessary volume of FastAmp Viral and Cell Solution is provided in the tube used for sample collection. All enzymatic components, reagents, and compatible buffers required for SARS-CoV-2 detection are included within Lyophilized Reagent Mix B and are reconstituted with Reagent Mix A (see protocol for details). A transilluminator or equivalent blue light emitting device is needed to visualize the assay results. A smartphone or smart device is necessary for automated interpretation of the SARS-CoV-2 detection results using the HandLens application.

Protocol materials

- FastAmp® Viral and Cell Solution for Covid-19 Testing Solution B **Catalog #**4633
- Screw cap tube 5 mL sterile **Thomas Scientific Catalog #**1188R46
- Reagent Mix A (In-tube SARS-CoV-2 resuspension mix)
- Lyophilized Reagent Mix B (In-tube SARS-CoV-2 detection mix)
- Screw cap tube 5 mL sterile **Thomas Scientific Catalog #**1188R46
- FastAmp® Viral and Cell Solution for Covid-19 Testing Solution B **Catalog #**4633
- Reagent Mix A (In-tube SARS-CoV-2 resuspension mix)
- Lyophilized Reagent Mix B (In-tube SARS-CoV-2 detection mix)
- FastAmp® Viral and Cell Solution for Covid-19 Testing Solution B **Catalog #**4633
- Screw cap tube 5 mL sterile **Thomas Scientific Catalog #**1188R46
- Reagent Mix A (In-tube SARS-CoV-2 resuspension mix)
- Lyophilized Reagent Mix B (In-tube SARS-CoV-2 detection mix)

Troubleshooting

Safety warnings

Please take care with potentially infectious sample material that does not come into contact with the provided viral lysis solution contained within the saliva sample collection tube.

Before start

Download the HandLens application on the user-provided smart device (smartphone, tablet, etc.). Clean workspace with disinfectant prior to starting the protocol.

Sample Collection and Viral Lysis

1 Expel approximately one drop of saliva into the sample collection tube and cap the tube.
Saliva collection tube contains necessary volume of FastAmp® Viral and Cell Solution.
Screw cap tube 5 mL sterile **Thomas Scientific Catalog #**1188R46

FastAmp® Viral and Cell Solution for Covid-19 Testing Solution B **Catalog #**4633

2 Mix saliva sample and FastAmp® Viral and Cell Solution by vortexing the closed sample collection tube for 00:00:05 .

3 Wait 00:05:00 , incubating sample at Room temperature , before proceeding to Step 4.

SARS-CoV-2 Detection

4 Pipette 15 µL of Reagent Mix A into a single uncapped well of the 96-well plate containing lyophilized Reagent Mix B. Mix by pipetting up and down gently.
Reagent Mix A (In-tube SARS-CoV-2 resuspension mix)

Lyophilized Reagent Mix B (In-tube SARS-CoV-2 detection mix)

5 Add 5 µL sample-viral lysis mix to Reagent Mix A and B well. Mix by pipetting up and down gently. Recap sample.

6 Wait 01:30:00 , incubating sample at Room temperature , before proceeding to Step 7.

In-tube Fluorescent Readout and Automated Analysis

7 Visualize the fluorescence of the sample using a transilluminator or equivalent blue light emitting device.

Equipment

NAME	BRAND	SKU
13 × 12 cm mini Transilluminator	Clare Chemical Research	DR22A

8 With the user-provided smart device such as a smartphone, open the HandLens application and select in-tube as the test type.

9 Take a photo of the plate, and select upload. The result of the test will appear on the smart device screen.