

Sep 08, 2020

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

Jon Arizti-Sanz^{1,2}, Catherine A. Freije¹, Chloe K. Boehm¹, Sameed M. Siddiqui^{1,3}, Allen M. Goodman¹, Tinna-Solveig F. Kosoko-Thoroddsen¹, A'Doriann Y. Bradley¹, Jeremy Johnson¹, Pardis C. Sabeti^{1,4,5,6,7}, Cameron Myhrvold^{1,8,7}

¹Broad Institute of Massachusetts Institute of Technology (MIT) and Harvard, Cambridge, MA 02142, USA.;

²Harvard-MIT Program in Health Sciences and Technology, 77 Massachusetts Ave., Cambridge, MA 02139, USA.;

³Computational and Systems Biology PhD Program, MIT, Cambridge, MA 02139, USA.;

⁴Harvard T.H. Chan School of Public Health, 677 Huntington Avenue, Boston, MA 02115, USA.;

⁵Department of Organismic and Evolutionary Biology, Harvard University, 26 Oxford Street, Cambridge, MA 02138, USA.;

⁶Howard Hughes Medical Institute, Chevy Chase, MD 20815, USA.;

⁷Massachusetts Consortium on Pathogen Readiness, Boston, MA, USA.;

⁸Department of Organismic and Evolutionary Biology, Harvard University, 26 Oxford Street, Cambridge, MA 02138, USA

Works for me dx.doi.org/10.17504/protocols.io.bk3rkym6



Catherine Freije

ABSTRACT

This protocol describes how to perform a SHINE in-tube fluorescent assay to detect SARS-CoV-2 RNA from a selfcollected 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).

DOI

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

KFYWORDS

CRISPR, SARS-CoV-2, nucleic acid diagnostic

LICENSE

This is an open access protocol distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited

IMAGE ATTRIBUTION

biorender.com

CREATED

Sep 08, 2020

LAST MODIFIED

Sep 08, 2020

DDOTOCOL INTECED ID

m protocols.io

09/08/2020

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 (09/08/2020). XPRIZE SHINE - In-tube Fluorescent SARS-CoV-2 Saliva Test. https://dx.doi.org/10.17504/protocols.io.bk3rkym6

41809

MATERIALS

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

STEPS MATERIALS

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

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

MATERIALS TEXT

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.

EQUIPMENT

NAME	CATALOG #	VENDOR
13 x 12 cm mini Transilluminator	DR22A	

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.

DISCLAIMER:

The protocol and its content is for informational and academic purposes only. It does not constitute legal, medical, clinical, or safety advice, or otherwise. Content added to protocols.io is not peer reviewed and may not have undergone a formal approval of any kind. Information presented in this protocol is not a substitute for independent professional judgment, advice, diagnosis, or treatment. Any action taken or not taken based on the protocol presented here is strictly at your own risk. You agree that none of the authors, contributors, administrators, nor anyone else associated with protocols.io, can be held responsible for your use of the information contained in or linked to this protocol.

BEFORE STARTING

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

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.

protocols.io
2
09/08/2020

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 (09/08/2020). XPRIZE SHINE - In-tube Fluorescent SARS-CoV-2 Saliva Test. https://dx.doi.org/10.17504/protocols.io.bk3rkym6

- Screw cap tube 5 mL sterile
 by 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.

protocols.io
3
09/08/2020

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 (09/08/2020). XPRIZE SHINE - In-tube Fluorescent SARS-CoV-2 Saliva Test. https://dx.doi.org/10.17504/protocols.io.bk3rkym6



- 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.