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

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1 Works for me

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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).

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KEYWORDS

CRISPR, SARS-CoV-2, nucleic acid diagnostic

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MATERIALS

NAME	CATALOG #	VENDOR
FastAmp® Viral and Cell Solution for Covid-19 Testing Solution B	4633	
Custom nasopharyngeal swab and collection tube		
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
Custom nasopharyngeal swab and collection tube		
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		

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 NP sample collection tube.

DISCLAIMER

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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

Open the nasopharyngeal (NP) collection tube and rotate the nasal swab (attached to the NP collection tube cap) 4 times around the inside of each nostril. Return the swab to the collection tube and cap the tube.

Nasopharyngeal collection tube contains necessary volume of FastAmp® Viral and Cell Solution.

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- Custom nasopharyngeal swab and collection tube
- FastAmp® Viral and Cell Solution for Covid-19 Testing Solution B
 Catalog #: 4633
- 2 Mix NP sample and FastAmp® Viral and Cell Solution by vortexing the closed sample collection tube for **© 00:00:05**.
- 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 🏚 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.

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- 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.