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FastAmp Saliva Room Storage Powder for SARS-CoV-2 (Covid19) Testing.

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1 *Works for me* This protocol is published without a DOI.

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SUBMIT TO PLOS ONE

ABSTRACT

FastAmp Saliva Room Storage Powder (Intact Genomics, Inc. Catalog #4607) enables the lysis of human cells and viral particles, while also stabilizing SARS-CoV-2 (COVID-19) RNA and human DNA/RNA in saliva. Utilizing this powder, storage at room temperature is possible for up to 10 days. The sample may then be directly tested with one step RT-qPCR kit for SARS-CoV-2 (COVID-19) detections (Intact Genomics, Inc. Catalog #4223) without RNA extraction steps. The test is used for screening purposes under class of "research used only" (RUO). This test is not for diagnostic purposes, but can be further developed to simplify diagnostic testing and other applications.

PROTOCOL CITATION

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KEYWORDS

SARS-CoV-2, Saliva, FastAmp

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GUIDELINES

This protocol is to be used for research use only.

SAFETY WARNINGS

Most institutions will require samples potentially containing full-length SARS-CoV-2 RNA to be handled in a biosafety level 2 cabinet. Please seek guidance from your local biosafety office on specific recommendations for working with samples which could contain live SARS-CoV-2 virus.

DISCLAIMER:

DISCLAIMER – FOR INFORMATIONAL PURPOSES ONLY; USE AT YOUR OWN RISK

The protocol content here is for informational purposes only and does not constitute legal, medical, clinical, or safety advice, or otherwise; content added to protocols.io is not peer reviewed and may not have undergone a formal

BEFORE STARTING

Refrain from consuming food or beverage (including water) for 30 minutes before providing a saliva sample.

1 Saliva can either collected independently by the individual or with the assistance of a healthcare worker or technician.

2 Before collection, clean hands using alcohol-based sanitizer or soap and water (no fragrance).

3 Ensure all collection materials are labelled with the correct identifying information.

4 Open the cap of the collection tube (5mL) containing FastAmp Saliva Room Storage Powder and gently expel 1mL saliva into the collection tube.

5 Once 1mL of saliva has been collected, securely close the cap of the collection tube.

6 Mix well until the powder is completely dissolved.

7 Sterilize the sample collection tube surface with 70% ethanol or a disinfecting wipe.

8 Store the sample collection tube at room temperature until transport to the laboratory for sample processing. The viral particles and human cells will be lysed by the powder reagents to release viral RNA and human DNA/RNA in the saliva mixture. Viral RNA and human DNA/RNA in the saliva mixture remains stable at room temperature for long term storage.

9 If proceeding to RT-qPCR, add 1ml Nuclease-free water into the sample collection tube (including collected 1mL saliva), total volume should be 2mL.

10 Vortex or swirl sample to mix until homogeneous.

11 Stand in a tube rack until precipitation of the viral particle/cell debris occurs (3 min).

12 Sample can be stored at room temperature (maximum~10 days).

13 The supernatant of the saliva mixture can later be used with one step RT-qPCR kit for SARS-CoV-2 (COVID-19) detections, without the need for RNA purification.

14 On ice, prepare a master mix for one step RT-qPCR.

Component	Volume
Supernatant of the saliva mixture	5µl
One step RT-qPCR 2x Master Mix	15µl
Primer/Probe mix (3.2nM/1.7nM)	3µl
Nuclease-free water	7µl

15 1Load the tubes into qPCR machine, and run the following thermocycler conditions

Step	Temperature	Time
1	42°C	30min
2	98°C	2min
3	98°C	10sec
4	56°C	30sec
5	Read plate	
Repeat steps 3-5 for 44 cycles.		

Real-time PCR (qPCR) instruments currently validated: Bio-Red CFX Connect.

16 Report results