RapiDxFire 1-step RT-qPCR System for SARS-CoV-2 detection quick protocol

This quick protocol is specific for SARS-CoV 2 detection using our <u>2019-nCoV CDC-</u> <u>qualified Probe and Primer Kits for SARS-CoV-2</u>. For general guidance on RT-qPCR reaction setups, please refer to our <u>RapiDxFire™ 1-step RT-qPCR System quick protocol</u>.

- 1. Thaw components at room temperature and mix well by vortex prior to use.
- Resuspend the 2019-nCoV CDC-qualified Probe and Primer Kits for SARS-CoV-2 oligonucleotides, following guidance in the <u>Centers For Disease Control and Prevention</u> (CDC) 2019-Novel Coronavirus (2019-nCoV) Real-Time RT-PCR Diagnostic Panel Instructions for Use.
- 3. Prepare reaction mixes (Table 1).

Component	1.6 µL*	5 µL	10 µL	20 µL	25 µL	Final concentration
RapiDxFire qPCR 5X Master Mix	0.32 µL	1 µL	2 µL	4 µL	5 µL	1X
EpiScript™ RNase <u>H- Reverse</u> <u>Transcriptase,</u> 200 Units/µL	0.04 µL	0.125 µL	0.25 µL	0.5 µL	0.625 µL	5 U/µL
2019-nCoV Probes and Primers	0.12 μL	0.375 µL	0.75 µL	1.5 μL	1.875 µL	1X
Template RNA	No more than 0.4 µL	No more than 1.25 μL	No more than 2.5 µL	No more than 5 μL	No more than 6.25 µL	As required
SuperROX (optional)	0.01 µL	0.03 µL	0.07 µL	0.13 µL	0.17 µL	100 nM
Water**	-	to 5 µL	to 10 µL	to 20 µL	to 25 µL	-

Table 1. RT-gPCR reaction mix set-up.

*If working with Array Tape[™] platforms from LGC, Biosearch Technologies[™] (e.g. IntelliQube[™], Nexar[™]), generation of a 2X mix is recommended for pipetting accuracy. Please contact <u>techsupport@lgcgroup.com</u> for further guidance.

**Volume to be adjusted accordingly to account for optional addition of passive reference dye.

 Place the reaction tubes/plates in a qPCR instrument and run the qPCR protocol. Ensure instrument is set to read at the appropriate channels for the selected probes (Table 2).

Step	Temperature	Time	Number of cycles	
1	42 °C	15 minutes	1 cycle	
2	95 °C	2 minutes	1 cycle	
3	95 °C	3 seconds	45 cycles	
3	55 °C	30 seconds		

Table 2. RT-qPCR thermal cycling condition.

For any queries about this quick guide, please contact techsupport@lgcgroup.com

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