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RNAPath Target Identification via Fluorescent Hybridization

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[1]Scooter Willis

In Development dx.doi.org/10.17504/protocols.io.bk37kyrn

XPRIZE Rapid Covid Testing RNAPath

Devin Willis

EXTERNAL LINK

http://rnapath.com/

DOI

dx.doi.org/10.17504/protocols.io.bk37kyrn

PROTOCOL CITATION

Devin Willis 2020. RNAPath Target Identification via Fluorescent Hybridization. **protocols.io** https://dx.doi.org/10.17504/protocols.io.bk37kyrn

EXTERNAL LINK

http://rnapath.com/

KEYWORDS

Molecular Instruments, COVID, Microscopy, Hybridization, Flourescence, RNA

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MATERIALS

NAME	CATALOG #	VENDOR
SSC (20X), RNase-free	AM9770	Thermo Fisher
Molecular Instruments HCR COVID Probe Set		Molecular Instruments
Molecular Instruments Alexa Flour 488 B1 HCR Amplifier		Molecular Instruments

STEPS MATERIALS

NAME	CATALOG #	VENDOR
Molecular Instruments HCR COVID Probe Set		Molecular Instruments
TE Buffer	12090015	Thermo Fisher

NAME	CATALOG #	VENDOR
SSC Buffer, 20X, 1L	V4261	Promega

BEFORE STARTING

Ensure you have cleaned the workspace with an appropriate RNAse away solution to prevent RNA degradation

Sample Preperation

1 Suspend RNA pellet in **2 µl TE Buffer**

TE Buffer
by Thermo Fisher
Catalog #: 12090015

Hybridization

2 Add **2 µl Molecular Instruments COVID Probe Set** to the RNA suspension

Molecular Instruments HCR COVID Probe Set
by Molecular Instruments
View

3 Place Molecular Instruments Alexa Flour 488 B1 HCR Amplifier in **90 °C** for **00:02:00**

4 Remove Molecular Instruments Alexa Flour 488 B1 HCR Amplifiers and place at **Room temperature** for **00:30:00**

5 Add **2 µl** of each Molecular Instruments Alexa Flour 488 B1 HCR Amplifier H1 and H2 to the RNA suspension

6 Dilute **100 µl** of 20x SSC Buffer to 5x SSC Buffer

SSC Buffer, 20X, 1L
by Promega
Catalog #: V4261

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7 Add **4 µl** of 5x SSC Buffer to the RNA Suspension

8 Incubate at **37 °C** for **12:00:00**

Imaging

9 Remove sample from incubation and pipette **1 µl** onto a glass slide with a coverslip

10 Image slide under a fluorescent microscope with filter cubes for FITC or Alexa Flour 488

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