Opentrons COVID-19 testing: Station A, Zymo kit, 24 or 48 samples

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[1]Opentrons Labworks

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Opentrons COVID-19 Testing

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Initial OT-2 setup

1   Clean the OT-2.

| 📄 | 📋 | **Cleaning an OT-2 COVID-19 Diagnostic Station**
by Max Marrone | PREVIEW | RUN | ∧ |

1.1   Wipe these parts of the OT-2 down with a 1:10 dilution of bleach:
1. The clear polycarbonate windows.
2. The black pipette stems. (Avoid the rest of the pipettes, including the ejectors.)
3. The aluminum deck.
4. The removable black trash bin.

1.2   Wait ⏱ **00:00:30** , then quickly rinse the bleach off with distilled water.

| 📄 | The aluminum on the OT-2 will be discolored if the bleach sits for too long. In the long term, it may also cause more serious corrosion. |

1.3   Wipe these parts of the OT-2 down with RNaseZap or RNase AWAY.

The same parts that you wiped down with bleach:
1. The clear polycarbonate windows.
2. The black pipette stems. (Avoid the rest of the pipettes, including the ejectors.)
3. The aluminum deck.
4. The removable black trash bin.

Plus these additional parts:
1. The bottoms of the pipette ejectors.
2. Any Temperature Modules or Magnetic Modules that the OT-2 has on its deck.
3. Any 96 well aluminum blocks that are going to be used on the OT-2.

1.4 Rinse the RNaseZap or RNase AWAY off with distilled water.

1.5 Wipe the OT-2 dry, or let the water evaporate.

Reagent preparation

2 Prepare the **reagent tube rack,** an Opentrons 24 tube rack.

2.1 Pipette **Proteinase K** prepared from the Zymo Quick-DNA/RNA Viral MagBead kit into a **1.5 mL snap cap tube.** The volume depends on how many samples are being processed:

Samples to process	Volume of Proteinase K (µL)
24	110 µL
48	220 µL

Place the Proteinase K tube in **well D1** of the **reagent tube rack.**

2.2 Pipette **internal extraction control RNA** prepared from the BP Genomics 2019-nCov Detection Assay kit into a **1.5 mL snap cap tube.** The volume depends on how many samples are being processed:

Samples to process	Volume of internal extraction control RNA (µL)
24	110 µL
48	220 µL

Place the internal extraction control RNA tube in **well D4** of the **reagent tube rack**.

OT-2 deck setup

3 Set up labware on the OT-2's deck.

3.1 In **deck slot 7,** place the **reagent tube rack.**

3.2 In **deck slot 3,** place a full, sterile **Opentrons 20 µL filter tip rack.**

3.3 In **deck slot 1,** place an empty, sterile **NEST 96 deep well plate.**

Running the OT-2

4 Run the 24- or 48-sample protocol on the OT-2.
- 24 samples: **StationA-24samples-Zymo-M-2020-04-13.py**
- 48 samples: **StationA-48samples-Zymo-M-2020-04-13.py**

Operating an OT-2 for COVID-19 testing
by Max Marrone

PREVIEW   RUN

4.1 Open the Opentrons App.

4.2 Ensure you are connected to the robot. In the **Robots** tab, you can try flipping the robot's lights on and off to test the connection.

4.3 Go to the **Run** tab.

Double-check the name at the top to make sure the correct protocol is uploaded.

4.4 Click **Start run**. The OT-2 will home its motors and then begin the protocol.

> ⚠ Do not click **Start run** more than once. If you do, a known bug will make the OT-2 run the protocol back-to-back.

> ⚠ **If something goes wrong and you need to abort the protocol:**
>
> 1. Shut down the OT-2 with the power switch on its back left side.
> 2. Turn the OT-2 back on. Wait a couple of minutes for the pipettes to rise.
> 3. Manually remove any tips attached to the pipettes. (This ensures that the pipettes will not aspirate liquid into themselves when they home.)
> 4. Reconnect to the OT-2 in the Opentrons App. Click the Home button to move the gantry out of the way so you can access the labware on the deck.

5 The output is the **NEST 96 deep well plate** in **deck slot 1.** It contains either 24 or 48 wells of 🧪**8 µl Proteinase K + internal extraction control RNA mix** .

Only the odd-numbered columnns (1, 3, 5, ...) are used. The even-numbered columns are skipped.