Station B: DNA Healthcare Solutions Extraction for COVID-19 Setup guide

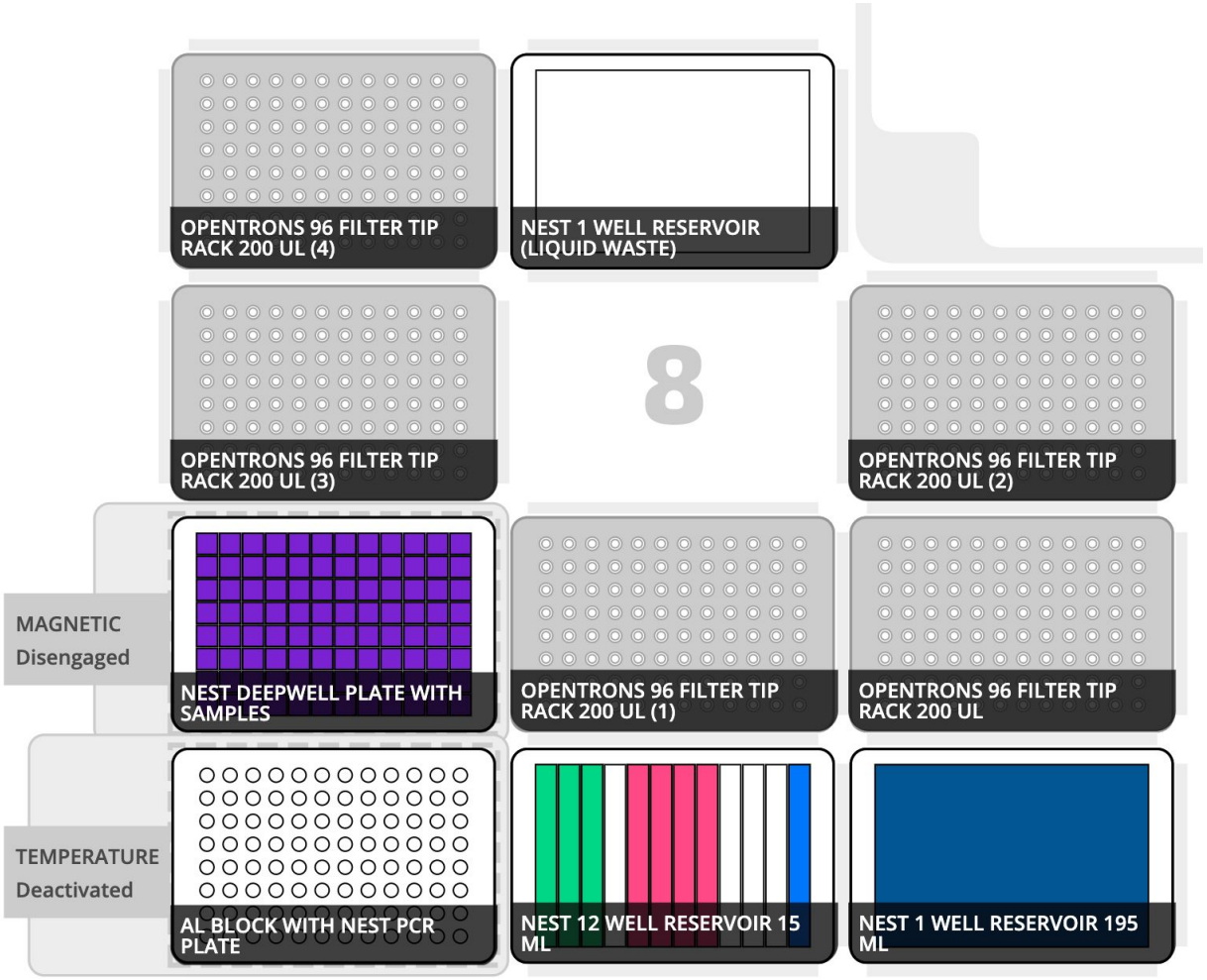
Code parameters:

- Change the sample number on line 11 (start at 8)
- Change the elution volume on line 13 (default is 100µL)
- Change the starting volume on line 14 (default is 320µL; 300µL of sample + 20µL of pk)

Pipettes:

- P300 multichannel on the left mount

Deck Layout:



Labware and module requirements:

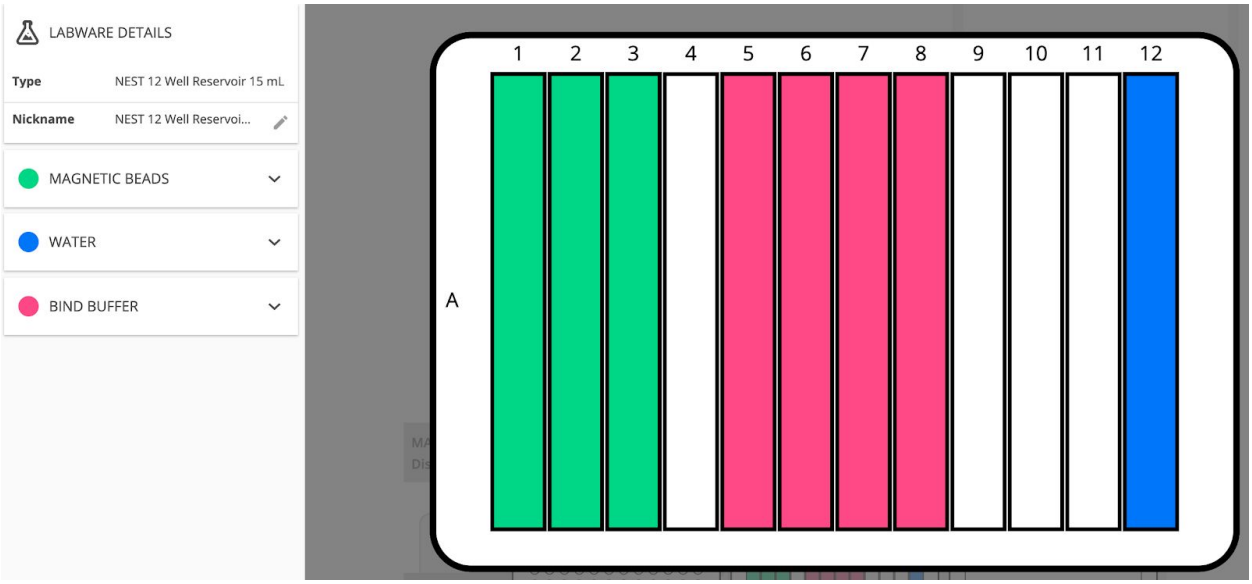
- 1 x Magnetic Module
- 2-5 x 200µL Tipracks
- 1 x NEST Deepwell Plate with Samples
- 2 x NEST 1-Well Reservoirs

- 1 x NEST 12-Well Reservoir
- 1 x NEST 96-Well PCR Plate
- 1 x Temperature Module with 96-Well Aluminum Block (optional)

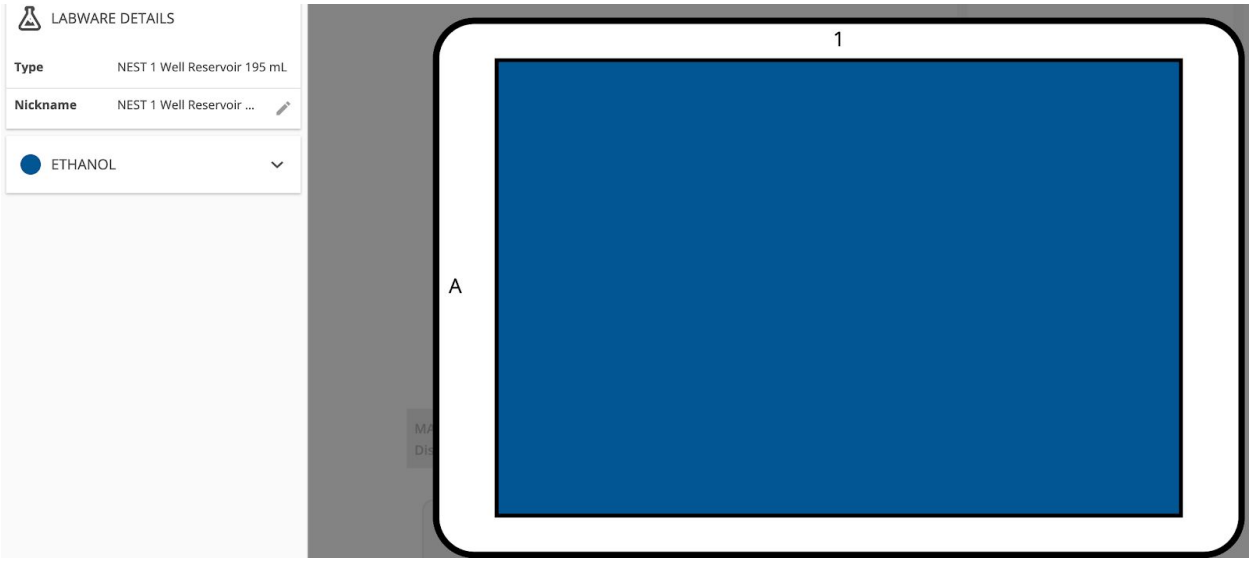
Volume requirements:
***Note**: the below volumes account for a 10% overage - the dead volume can be adjusted depending on the calibration of the pipette to the labware, but it's recommended to have an overage of about 10%*

Mastermix components	Volume per sample (µL)	Volume for 8 samples (mL)	Volume for 24 samples (mL)	Volume for 48 samples (mL)
Magnetic Beads	300	2.6	7.8	15.6
Bind Buffer	450	4	12	24
Wash Buffer	1000 (x3)	26	78	156
Nuclease-Free Water	100	1	3	5.5

Slot 2, NEST 12-Well Reservoir Setup -
Magnetic Beads - Slots 1-3, each slot can accommodate up to 4 columns of samples.
Bind Buffer - Slots 5-8, each slot can accommodate up to 3 columns of samples.
Nuclease-Free Water - Slot 12



Slot 3, NEST 1-Well Reservoir Setup -
Load with wash buffer (70% alcohol) to accommodate 3 washes per sample



Before you begin:

1. Pre-cool the Temperature Module in the Opentrons App to 4°C, if using.
2. Add tipracks. The tiprack in slot 6 will always be used to add reagents. The other tipracks will be accessed in this order: slot 5, 9, 7, 10 and will be used for mixing and removing supernatant. Each column of samples will need 4 columns of tips for mixing and removing supernatant (ie, if running 8 samples, only 4 columns of tips from slot 5 will be used; if running 16 samples, 8 columns of tips from slot 5).
3. Add reservoirs for reagents (slots 2, 3) and fill with respective reagents; add an empty reservoir for liquid waste in slot 11 (this setup can accommodate an extra reservoir for liquid waste in slot 8, if needed).
4. Add the NEST deepwell plate with samples (from Station A) on top of the magnetic module in slot 4.
5. Add the elution plate to slot 1 (on top of the temperature module with aluminum block, if using).

This protocol utilizes the multi-channel pipette and will be transferring elutions from the deepwell plate containing samples (slot 4) to the elution plate (slot 1) for RT-qPCR in a 1-to-1 transfer (A1 → A1, B1 → B1, etc.). In the case that the sample number is not divisible by eight, the protocol will still fill the entire column of the elution plate, but the wells that did not contain samples, will contain nuclease-free water.