Update: June 6, 2020

Station B: Chemagic Viral 300 DNA/RNA Kit - Perkin Elmer

This protocol starts immediately after lysing the sample and starts at step 2 with the addition of magnetic beads and binding buffer 2

Code parameters:

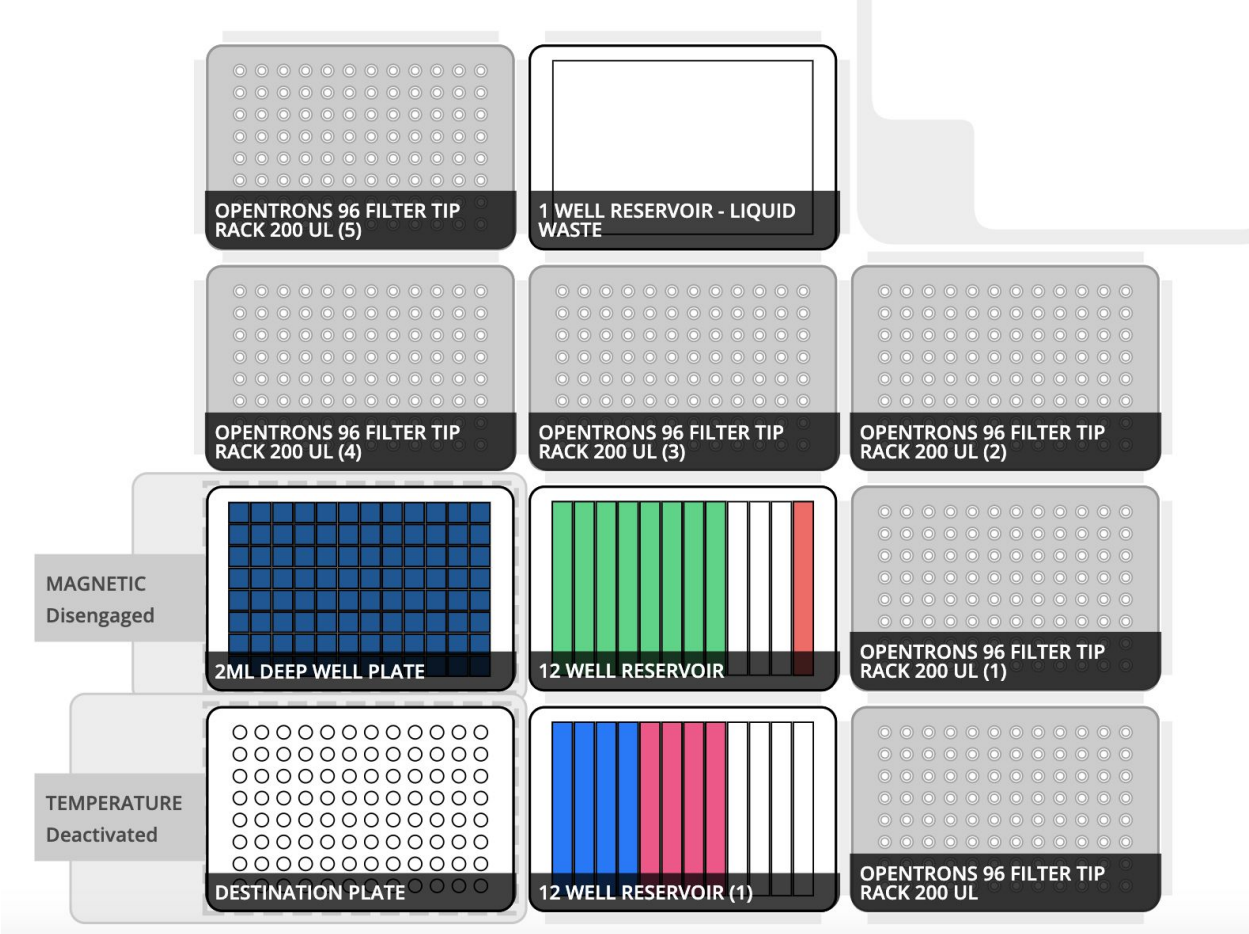
- Change the sample number on line 14 (default is 8, max is 94)
- Change the elution volume on line 15 (default is 50µl, max is 100µl)
- Change the sample starting volume on line 16 (default is 620µl)
- Tip rack tracking can be changed from False to True on line 17 (default is False)
- Make “tiprack parking” True or False on line 18* (default is True)

**if you selected True for “tiprack parking,” tips used for the same buffers with the same samples will be reused where 1 tiprack turns into a tiprack where used tips are “parked”. This method has low risk of contamination and is highly recommended to avoid pauses to refill tipracks.*

Pipettes:

- P300 multichannel on the left mount

Deck Layout:



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Labware and module requirements:

- 1 x magnetic module
- 1 x temperature module
- 1 x 2mL deep well plate **[input with lysates]**
- 6 x 200µl filter tip racks (10 x 200µl if you select false for tiprack parking)
- 1 x 1 well reservoir **[loaded empty for the liquid waste in slot 11]**
- 2 x 12 well reservoir with reagents **[1 x holds Magnetic Beads & Binding Buffer 2 Mixture, and Elution Buffer 6, 1 x holds Wash Buffer 3 and Wash Buffer 4]**
- 1 x 96 well aluminum block loaded on top of the temperature module in slot 1
- 1 x 96 well PCR plate ***or*** PCR strip tubes to match the number of samples **[output with eluates/extractions]**

Volume requirements:

Note*: the below volumes account for a dead volume - the dead volume can be adjusted depending on the calibration of the pipette to the labware, but it's best to have a dead volume of at least 10%*

Reagents	**Volume per sample (µl)**	**Volume for 8 samples (µl)**	**Volume for 48 samples (mL)**	**Volume for 96 samples (mL)**
Magnetic Beads & Binding Buffer 2	1000	12,000	52	100
Wash Buffer 3	500	6,000	26	50
Wash Buffer 4	500	6,000	26	50
Elution Buffer 6	50 - 100	600 - 1,200	2.6 - 5.2	5 - 10

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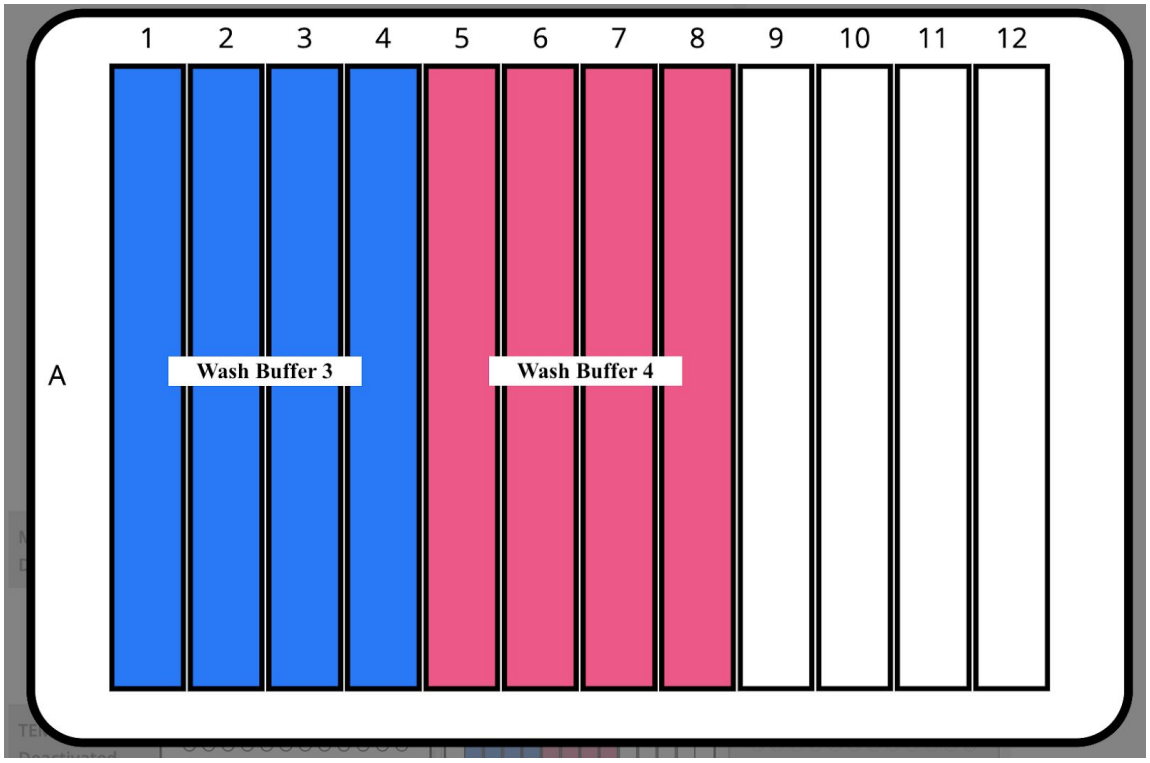
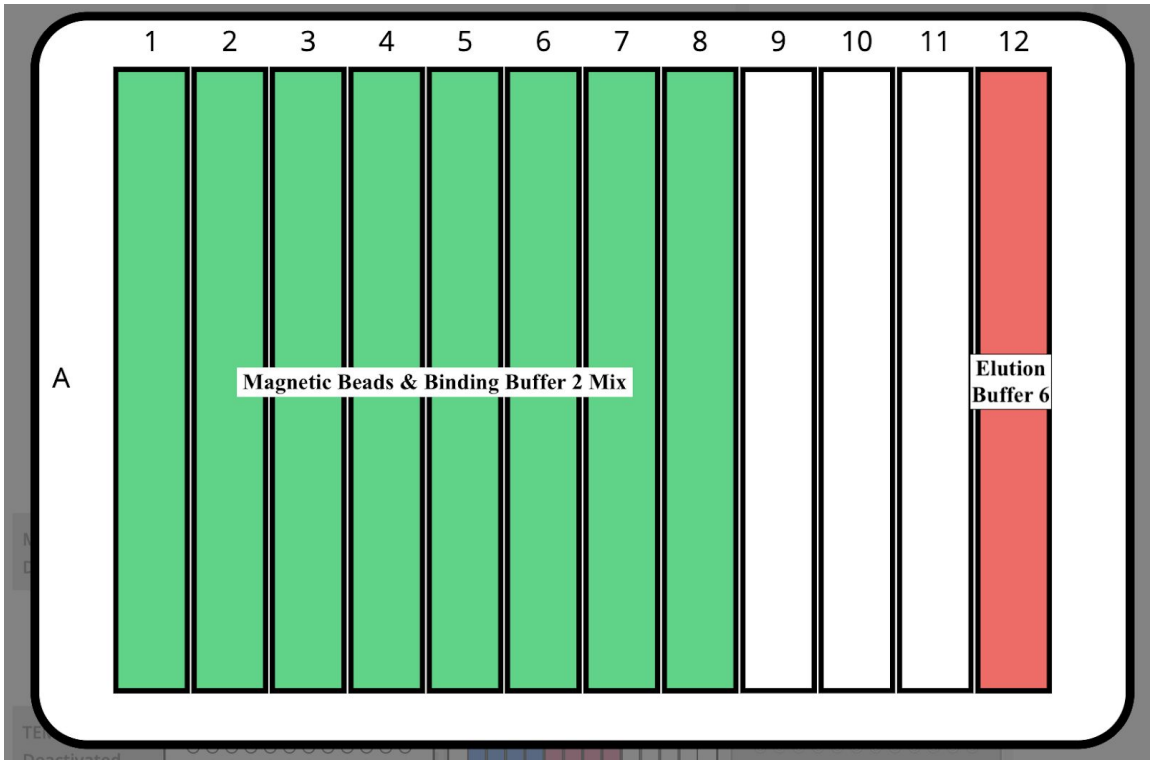
Slot 2 - 12 well reservoir setup



Slot 5 - 12 well reservoir setup



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Before you begin:

1. Pre-cool the Temperature Module in the Opentrons App to 4°C
2. Create the **Magnetic Beads & Binding Buffer 2 Mixture**

	Volume per sample	**Volume for 8 samples**	**Volume for 48 samples**	**Volume for 96 samples**
Magnetic Beads	20 µl	240 µl	1,040 µl	2 mL
Binding Buffer 2	980 µl	11,760 µl	50.9 mL	98 mL

Note: *the beads settle quickly so be sure to vortex the solution thoroughly before adding the mixture to the reservoir*

3. Add the Wash Buffer 3 and Wash Buffer 4 to the 12 well reservoir in slot 2
4. Add the Magnetic Bead & Binding Buffer Mixture and Elution Buffer 6 to the 12 well reservoir in slot 5
5. Place the deep well plate filled with lysates on top of the magnetic module in slot 4.
6. Add a 96 well aluminum block and the 96 well PCR plate or PCR strip tubes on top of the Temperature Module in slot 1

The final plate of eluates/extractions will be found on top of the temperature module in slot 1. Once the run is complete, please proceed to Station C for RT-PCR set up.