

## **Quick-Start Protocol**

## CarbonPrep Wastewater - Manual Protocol

Shelf-life

- Carbon beads have a shelf-life of at least two years.
- RNA pre-binding solution (RPS), RPS wash buffer (RPS WB), wash buffer (WB) and Binding buffer (BB) have a shelf life of one year.

Further Information

- OT-2 FAQ Sheets: See product page. Safety Data Sheets: See product page.
- Technical assistance: support@magnetics.life

Before Starting

- Add ethanol to bottles which require it. The amount to add is listed on the bottle.
  a. After adding ethanol, mark the bottle in the space provided on the label.
- 2. The complete kit will have seven solutions, RPS Wash buffer (RPS), Wash Buffer (WB), Carbon beads, and elution buffer (EB).

Protocol

- 1. To 20-50 mL of wastewater add 70uL of carbon beads.
- 2. Gently stir for 2-4 hours. We recommend a mixer such a Rotator Genie like this one found on <u>amazon</u>. The beads should look clumpy as shown below.



Image 1: Wastewater sample and LM carbon beads in RPSWB after tumbling for 4 hours.

- a If they do not look clumpy, consider increasing the volume of wastewater. The sample will yield results even if it doesn't look clumpy, but a higher volume of wastewater may be optimal.
- 3. Separate magnetic beads with a magnet and remove liquid.
- 4. Re-suspend beads in 750uL of RPS wash buffer and transfer to a 1.5 ml tube.
- 5. Place 1.5 ml tube on a magnetic stand. Wait for the beads to separate and remove the liquid.
- 6. Re-suspend with 750uL of Wash buffer and transfer to a clean 1.5mL Eppendorf tube.

- a It is recommended to move this solution to a new tube to decrease contamination.
- 7. Remove liquid after bead separation.
- 8. Re-suspend with 500uL of Wash buffer and decant liquid after bead separation.
- 9. With the liquid removed, allow the beads to dry at room temperature for 10 minutes.
- 10. Re-suspend the beads with 40-80uL of elution buffer. Heat samples in a water bath to 65 C for 10 minutes.
- 11. Centrifuge 10,000 x g for 2 minutes to pellet carbon beads. You can also use the magnetic stand, but centrifuging will compact the beads which will maximize recovery.
- 12. Place the sample on a magnetic stand. Remove the clear liquid and place it into a fresh 1.5mL tube. This is your sample.