SPINeasy™ DNA Kit for Water

Cat. No.: 116536050 (50 PREPS) / 116536000 (5 PREPS)

Quick-Start Protocol

Revision August 20, 2024

Scan QR code for more information from instruction manual

Notes before starting:

- Filter Membrane for collection of microorganisms is provided in the kit.
- If Lysis Buffer W1 has precipitated, heat at 55 °C to dissolve precipitate.
- Add 30 mL (3 mL for sample kit) isopropanol to Binding Buffer W and mark on the bottle.
- Add 21 mL (2.1 mL for sample kit) absolute ethanol to Wash Buffer W1 and mark on the bottle.
- Add 50 mL (5 mL for sample kit) absolute ethanol to Wash Buffer W2 and mark on the bottle.
- Centrifugation speed stated in the manual will be a guideline; use the maximum speed available if 14,000 *g* is not feasible.

Column preparation

1. Place **Column W1** on top of **2.0 mL Collection Tubes** (provided) , add **200 µL Equilibration Buffer** to Column W1 to ensure its performance. Centrifuge for **1 min @ maximum speed**. Discard the flow-through and reuse the collection tube.

Lyse

2. Refer to the back of this quick-start protocol for **Filter Membrane** preparation and usage. After placement of filter membrane, proceed to the next step.
3. Add **980 µL Lysis Buffer W1, 120 µL Lysis Buffer W2,** and **10 µL RNase A** to the sample in the **Lysing Matrix E** tube. Homogenize using a Fastprep® at 6 m/s for 30s for 2 cycles, pause for 2 minutes between cycles with a Metal MidiPrep™ 5 mL metal adaptor (Cat. No. 116002544) or vortex for **10 min @ maximum speed**.

Purify

4. Add **250 µL Inhibitor Removal W** to the Lysing Matrix E tube, mix by inverting the tube **20 times**.
5. Centrifuge for **5 mins @ 5,000 *g*** to pellet precipitate. Transfer up to **900 µL** supernatant to a clean 2 mL microcentrifuge tube (not provided).

Bind

6. Add up to **900 µL** (or equal volume) **Binding Buffer W** to the supernatant. Vortex to mix.
7. Transfer **800 µL** of the mixture to Column W1. Centrifuge for **30 secs @ 14,000 *g***. Empty the collection tube. Repeat the process once.

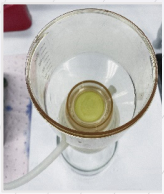
Wash

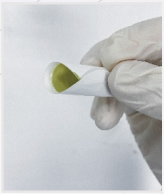
8. Add **500 µL Wash Buffer W1** to Column W1. Centrifuge for **30 secs @ 14,000 *g***. Empty the collection tube.
9. Add **500 µL Wash Buffer W2** to Column W1. Centrifuge for **30 secs @ 14,000 *g***. Empty the collection tube. Repeat the wash process with **Wash Buffer W2**.
10. Without addition of any liquid, centrifuge for **2 mins @ 14,000 *g*** to dry the column.
11. Discard the collection tube, place Column W1 on top of **1.5 mL Collection Tubes** (provided). Air dry the column for **5 mins** at room temperature.

Elute

12. Heat **DES Buffer** to 55 ˚C using a water bath while waiting.
13. Add **100 µL** pre-heated **DES Buffer** to the center of column.
14. Centrifuge for **1 min @ 14,000 *g*** to elute DNA.
15. Eluted DNA is now ready for downstream applications. Store at -20 ˚C or -80 ˚C for extended periods.

Filter Membrane Preparation and Usage




Preparation	Filtration	Filter Removal	Rolling	Placement
Setting up vacuum filter set	Filter highest possible amount of sample	Using forceps, pick up the filter membrane	Roll filter into cylinder shape with top side (microbes trapping side) facing inwards	Insert filter into Lysing Matrix E tube

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