# SPINeasy™ DNA Maxi Kit for Soil

Cat. No.: 116549010 (10 PREPS)

## **Quick-Start Protocol**

#### Revision Apr 2023

# WP®



#### Scan QR code for more information from instruction manual

### Notes before starting:

- · Store Buffer MS2 at 2-8 °C upon receiving the kit.
- If Buffer MS1B has precipitated, heat at 37 °C until precipitate dissolves.
- · All the centrifugations must be done using a swinging bucket able to reach at least
- 3,000 x g. Higher speed is preferred.
- · It is advised to pre-position the plasticwares and columns used during the extraction as depicted in the next page.

1. Add up to 10 g (high biomass) or 20 g (low biomass) of soil sample to the **50 mL BigPrep Lysing Matrix YB.** Add 5 mL of **Equilibration Buffer** to **Column MS**. To be efficient, the Column MS need to be incubated with the Equilibration Buffer for at least 5 min. Spin for 2 min @ maximal speed.

2. Add 15 mL of **Buffer MS1A** and 3 mL **Buffer MS1B** (high biomass soil) or 12 mL of Buffer MS1A and 6 mL Buffer MS1B (low biomass soil). Invert the tube 5X to mix and homogenize using Fastprep 5 m/s, 45 s, BigPrep™ adapter, or vortex for 20 min @ 2500-2800 rpm, Spin for 10 min @ maximal speed.

3. Pour the supernatant into a new 50 mL Tube (provided) without disrupting the pellet. Add 2 mL of **Buffer MS2**, shake the tube 5X. Spin for 8 min @ maximal speed .

Note: The volume of Buffer MS2 is suitable for most soils. Up to 4 mL of Buffer MS2 may be used for soil with high humic acid content while 0.5 mL can be used for low biomass/contaminant soil. The volume may need to be optimized by the user as an excess of Buffer MS2 may decrease the nucleic acid yield.

4. Transfer up to 15 mL of supernatant into a new 50 mL Tube (provided) without disrupting the pellet. 
Optional: A 100  $\mu$ m sterile strainer (not provided) can be used to filter floating particles if any. 
Add 18 mL of Buffer MS3. If more supernatant is transferred, add 1.2 volume of Buffer MS3 instead. Shake the tube 5X. The subsequent steps can be performed using either centrifuge or vacuum manifold (faster).

#### Centrifuge

Add ~15 mL of the lysate into the Column MS. Avoid transferring soil particles if any. Spin for 2 min @ maximal speed. Discard the flow-through and place the column back into the collection tube. Repeat once.

#### 5. First Wash

Optional: If soil particles are found on the membrane, add 2-5 mL of **Buffer MS4** to the column, swirl the column and discard the buffer. Repeat if required.

-Add 10 mL of Buffer MS4 and spin for 2 min @ maximal speed. Discard the flow-through and place the column back into the same collection tube.

#### Vacuum manifold

Transfer the Column MS into the vacuum manifold's luer connector. The collection tube will be used in the next. Load ~15 mL of the lysate into the Column MS while avoiding transferring soil particles if any. Apply vacuum. Repeat until all the lysate has been loaded.

#### 5. First Wash

Optional: If soil particles are found on the membrane, add 2-5 mL of **Buffer MS4** to the column, swing the column and discard the buffer. Repeat if required.

-Add 10 mL of Buffer MS4 and apply vacuum. Transfer back the column to its collection tube.

- 6. Second Wash. Add 15 mL **Buffer MS5** and spin for 2 min @ maximal speed. Transfer the Column MS into a new 50 mL Tube (provided).
- 7. Column drying. Spin for 8 min @ maximal speed. If soil particles are still observed on the membrane, invert the column, and tap the column against the bench to discard the debris.

8. Transfer the column to a new 50 mL Tube (provided). Add 2 mL of **Elution Buffer** to the Column MS membrane. Incubate for  $\geq 2$  min, spin for 1 min @ maximal speed and reload again the eluted DNA. Wait  $\geq 5$ min and spin for 4 min @ maximal speed. The expected elution volume is  $\sim 1.8$  mL.

Note: For maximal yield, the second elution may be performed with 1-3 mL of fresh Elution Buffer instead. The genomic DNA can be readily used for downstream application. For extended storage, add **Buffer MS6** provided at 200X concentration to obtain DNA resuspended in 5 mM Tris and 0.1 mM EDTA (i.e.,  $\sim$ 9  $\mu$ L for 1.8 mL of DNA).

Equilibrate and Homogenize

펄

Vash

Elute

Cat. No.: 116549010 (10 PREPS)



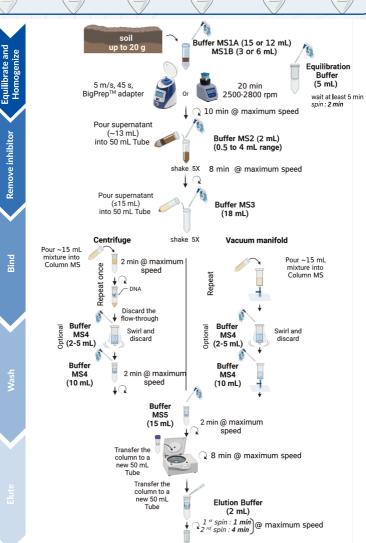
## **Quick-Start Protocol**

Revision Apr 2023

Homogenization Equilibration Inhibitor removal

Sw umm WS

Sw umm





Flow chart

**Plasticwares**