MagListo™ 5M Plant Genomic DNA Extraction Kit (K-3605)

Ⅰ Before You Begin

1) Completely dissolve one vial of **Proteinase K** in **1,250 µl** of nuclease free water. For short term storage, dissolved Proteinase K should be stored at **4°C**. For long term storage, it is recommended to aliquot the enzyme into separate tubes and store at **-20°C**.
2) Completely dissolve one vial of **RNase A** in **600 µl** of nuclease-free water. For short term storage, dissolved RNase A should be stored at **4°C**. For long term storage, it is recommended to aliquot the enzyme into separate tubes and store at **-20°C**.
3) Add correct amount of absolute ethanol to PWM1 Buffer.

Ⅱ DNA Extraction from Plant

1) Disrupt (or homogenize) the **tissue (~100 mg) or seed (~50 mg)** then place them into a 1.5 ml tube.
2) Add **300 µl** of **PL Buffer** and **10 µl** of **RNase A solution**, and mix thoroughly by vortexing.
3) Add **20 µl** of **Proteinase K**, mix thoroughly by vortexing and incubate at 60℃ for 10 min.
4) Add **100 µl** of **PC Buffer**, mix thoroughly by vortexing and incubate for 5 min on ice.
5) Centrifuge the tube at 13,000 rpm for 5 min.
6) Take the supernatant only and transfer into a new 1.5 ml tube.
7) Add **2 volumes** of **PWM1 Buffer** to the clear lysate, and mix thoroughly by vortexing.
8) Add **100 µl** of **Magnetic NanoBead solution**, and mix thoroughly by vortexing.
(Note) Please vortex Magnetic Nanobead Solution well before use.
9) Place the tube in ***MagListo*™-2 Magnetic Separation Rack** with the magnet plate attached and invert the rack gently 3~4 times.
10) Without removing the tube from *MagListo*™ rack, remove the supernatant.
11) Detach the magnet plate from *MagListo*™ stand. Add **500 µl** of **PWM1 Buffer**. Mix thoroughly vortexing.
12) Attach the magnet plate to *MagListo*™ stand and invert the tube 3~4 times gently until the beads bind tightly to magnet.
13) Without removing the tube from *MagListo*™ rack, remove the supernatant.
14) Detach the magnet plate from *MagListo*™ stand. Add **700 µl** of **W2 Buffer**. Mix thoroughly by vortexing, and repeat the above step 12~13.
15) Without removing the tube from *MagListo*™ rack, add **700 µl** of **WE Buffer** to "the opposite side of bead". Close the cap and gently invert the tube twice.
16) Discard the supernatant and completely remove the remaining supernatant by blotting.
17) Detach the magnet plate from *MagListo*™ stand. Add **100 µl** of **EA Buffer**. Mix thoroughly by vortexing and incubate at 60℃ for 1 min.

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18) Attach the magnet plate to *MagListo*™ stand and invert the tube 3~4 times gently until the beads bind tightly to magnet.
19) Without removing the tube from *MagListo*™ rack, transfer the supernatant containing DNA carefully to a sterile microcentrifuge tube.

※ For more information, please visit www.bioneer.com and refer to the User's Guide of this kit.

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Before You Begin

1) Completely dissolve **Proteinase K** in **1,250 µl** of nuclease free water.
2) Completely dissolve **RNase A** in **600 µl** of nuclease free water.
3) Add correct amount of **absolute ethanol** to **PWM1 Buffer**.
4) Disrupt (or homogenize) the plant sample.

Step	Image	Description
Lysis		Add Reagents to the sample and mix them completely - Sample : **~100 mg** of Tissue **~50 mg** of Seed - **PL Buffer : 300 µl** - **RNase A : 10 µl** - **Proteinase K : 20 µl**
Incubation		60°C Heating block for 10 min
Precipitation		Add **PC Buffer 100 µl** and mix
Incubation		Ice for 5 min
Precipitates removal		Centrifuge at 13,000 rpm for 5 min
DNA precipitation		Take the supernatant into a new tube. Add **PWM1 Buffer 2 volumes** to the supernatant and mix (ex. Supernatant 300 µl, PWM1 600 µl)
Binding		Add **Magnetic NanoBead 100 µl** and mix until the beads are fully resuspended
		Follow these 3 Magnet Attach/Detach Step 1. Attach magnet 2. Discard the supernatant 3. Detach magnet
1st Wash		Add **PWM1 Buffer 500 µl** and mix until the beads are fully resuspended
		Repeat the above magnet attach/detach step (step **1, 2 and 3**)
2nd Wash		Add **W2 Buffer 700 µl** and mix until the beads are fully resuspended
		Repeat the above magnet attach step (step **1 and 2**)
3rd Wash		Add **WE Buffer 700 µl** to "the opposite side of the bead pellet" Close the cap and gently invert the rack twice
		Repeat the above magnet detach step (step **2 and 3**)
Elution		Add **EA Buffer 100 µl** and mix
		60°C Heating block for 1 min