

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°C** 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°C** for 1 min.





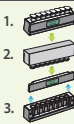




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- 18) Attach the magnet plate to *MagListo™* stand and invert the tube 3~4 times gently until the beads bind tightly to magnet.
- 19) Without removing the tube from *MagListo™* rack, transfer the supernatant containing DNA carefully to a sterile microcentrifuge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

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- 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
1 st 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)
2 nd Wash		Add W2 Buffer 700 µl and mix until the beads are fully resuspended
		Repeat the above magnet attach step (step 1 and 2)
3 rd 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