

## Kit Description

CatchGene™ Plant DNA Kit is a very broad spectrum kit. It's able to purify gDNA from a variety of plant samples such as rice, tobacco, Arabidopsis thaliana, Dendrobium nobile, corn, Pachira aquatica, evergreen, Epipremnum aureum... etc, also can extract gDNA from fungi. PL1 buffer has a good lytic effect to effectively lyse plant cell wall. When it combined with a powerful PL2 buffer, they could remove plant-specific interference and inhibitors such as polysaccharides and polyphenols. Nucleic acid can be adjusted to align with the column tube, the best combination state. CatchGene Plant DNA Kit allows the extraction of high-yielding and high-purity nucleic acids.

## Kit Content

	4 rxn	50 rxn	250 rxn	
Spin Column	4	50	250	pcs
Collection Tube (2 ml)	12	150	750	pcs
Buffer PL1	1.92	24	120	ml
Buffer PL2	0.63	7.8	39	ml
Buffer PL3 (concentrated)	0.96	12	60	ml
Buffer W1 (concentrated)	1.68	21	105	ml
Buffer W2 (concentrated)	0.68	8.4	42	ml
Elution Buffer	0.96	12	60	ml

## Kit Storage

Buffer, solvent and consumables, please store at 15-25 °C.

## Kit Preparation

- 1. Prepare Buffer PL3**  
Add two volume of 100% EtOH into Buffer PL3 (concentrated) to get Buffer PL3.  
After adding 100% EtOH, please check the sticker on the bottle and close the cap tightly.
- 2. Prepare Buffer W1**  
Add equal volume of 100% EtOH into Buffer W1 (concentrated) to get Buffer W1.  
After adding 100% EtOH, please check the sticker on the bottle and close the cap tightly.
- 3. Prepare Buffer W2**  
Add 4 volume of 100% EtOH into Buffer W2 (concentrated) to get Buffer W2.  
After adding 100% EtOH, please check the sticker on the bottle and close the cap tightly.

## Sample Pretreatment

### Pretreat of plant and fungi samples (fungi, corn, tobacco, rice, wheat...etc.)

1. Weigh 50mg (no more than 100mg) fresh or frozen plant samples ; or 10mg dried plant samples.  
(In the case of plants or fungi growing on the medium, heat to 80 ° C, remove the medium and rinse with PBS for several times, remove the liquid as possible and weigh.)
2. Please use the following method to homogenize the plant tissue.
  - A. Liquid nitrogen dry homogenization
    - a. Homogenize using a tissue homogenizer (such as Tissue Lysser, Beads Beater...etc.)
      1. First of all, try to cut the sample into small pieces (2mm x 2mm), then place in a shockproof 2ml sample tube.
      2. Add the appropriate amount (1-3 beads) and the appropriate size (3-5 mm) of acid-base resistance stainless steel beads.
      3. Immerse the sample tube in liquid nitrogen, to completely cool down the sample, the sample tube and the steel beads (at least 2 minutes).
      4. Put the sample into homogenizer, set the frequency 30 Hz for 30 seconds.
      5. Take out the sample tube to check if the sample is homogenized into powder and put it into liquid nitrogen for storage as soon as possible.
      6. If the sample is not homogeneous, repeat steps 3-5 to completely homogenize the sample to powder.
      7. Add 400 Buffer PL1 and vortex vigorously for 30 seconds.
    - b. Use a mortar and pestle for homogenization
      1. Pour some liquid nitrogen to cool the mortar and pestle.
      2. Place the sample in a mortar.
      3. Pour some liquid nitrogen in the mortar and grind the sample to powder.
      4. Move the powder sample into a sample tube which has been pre-cooled with liquid nitrogen.
      5. Add 400 Buffer PL1 and vortex vigorously for 30 seconds.

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## B. Lysate wet homogenization

### a. Homogenize using a tissue homogenizer (such as Tissue Lysser, Beads Beater...etc.)

1. First of all, try to cut the sample into small pieces (2mm x 2mm), then place in a shockproof 2ml sample tube.
2. Add the appropriate amount (1-3 beads) and the appropriate size (3-5 mm) of acid-base resistance stainless steel beads.
3. Add 400 Buffer PL1 and use a tissue homogenizer to homogenize.

## General Protocol

1. Place the plant tissue homogenate (containing 400  $\mu$ l Buffer PL1) at 65 ° C for 10-20 minutes (vortex every 3-5 minutes).
2. Add 130  $\mu$ l of Buffer PL2, vortex vigorously for 30 seconds and react on ice for 5 minutes.
3. Centrifuge at 11,000 x g for 5 mins.
4. Take the supernatant to a 1.5 ml tube (avoid any plant tissue fragments).
5. Add 1.5 times the supernatant volume of Buffer PL3 to a 1.5 ml tube (e.g. add 600  $\mu$ l of PL3 buffer to 400  $\mu$ l of the supernatant ) and mix thoroughly by pipetting.
6. Transfer 700  $\mu$ l lysate to the Spin Column (with 2ml Tube), centrifuge at 11,000 x g for 1 min, discard the flow-through.
7. Transfer the rest lysate to the Spin Column (with 2ml Tube), centrifuge at 11,000 x g for 1 min.
8. Carefully move the Spin Column to a new Collection Tube (2 ml).
9. Add 700  $\mu$ l Buffer W1 into the Spin Column, centrifuge at 11,000 x g for 1 min, discard the flow-through.
10. Add 700  $\mu$ l Buffer W2 into the Spin Column, centrifuge at 11,000 x g for 1 min, discard the flow-through.
11. Carefully move the Spin Column to a new Collection Tube (2 ml), centrifuge at full speed (~18,000 x g) for 3 min.
12. Place the spin column into a new 1.5 ml micro-centrifuge tube (not provided), add 50-200  $\mu$ l Elution Buffer and incubation at room temperature for 3 mins.
13. Centrifuge at 11,000 x g for 1 min for elution.

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