

# Plant DNA Extraction Kit

## Kit Contents:

| Components                 | 50 reactions |
|----------------------------|--------------|
| PB ( Blood Binding Buffer) | 12 ml        |
| PW1 (Wash Buffer)          | 15 ml        |
| PW2 (Wash Buffer)          | 12 ml        |
| PE (Elution Buffer)        | 12 ml        |
| PK (PK Storage Buffer)     | 1 ml         |
| Proteinase k               | 20 mg        |
| Spin Column                | 50 pcs       |
| Collection Tube            | 2 x 50 pcs   |

## Before Starting

1. Add 10 ml of absolute ethanol to the PW1 (only at the first use).
2. Add 48 ml of absolute ethanol to the PW2 (only at the first use).
3. Add Proteinase K (PK) solution to the lyophilized powder of proteinase K and store at -20 °C until usage (only at the first use).
4. Check PW1 and PB for salt precipitation. Redissolve any precipitation at 50 °C.
5. Preheat the solution of PE to 56 °C before starting the extraction process to enhance DNA extraction yield.

## Protocol:

1. Cut off 50-100 mg of fresh or frozen plant tissue or 50 mg of dry sample. Freeze the sample with liquid nitrogen. Grind the sample to a fine powder then transfer it to a 1.5 ml micro tube by adding 200 µl PB. Add 20 µl of proteinase K, and finally 200 µl of PB into a 1.5 ml micro tube.
2. Mix them well by vortexing (15 s) and incubate at 57 °C for 30-60 min, depending on sample. Spin for 1 min at 13000 rpm to remove debris and transfer supernatant to the new tube.
3. Add 200 µl of absolute ethanol and mix it by vortexing (10 s).
4. After a quick spin, carefully transfer lysate to the spin column. Do not touch upper rim of column. Spin for 1 min at 13000 rpm.
5. Replace the collection tube with a new one.
6. Add 500 µl of PW1 and spin for 1 min at 13000 rpm.
7. Pour off the flow-through of collection tube.
8. Add 700 µl of PW2 and spin for 1 min at 13000 rpm.
9. Pour off the flow-through of collection tube.
10. Repeat step 8 and 9 with 500 µl of PW2 (optional)
11. Spin for 1 min at 13000 rpm to remove the remaining of the wash buffer. Transfer the spin column to a new 1.5 ml micro tube.
12. Add 100 µl of preheated PE, wait 3 min at room temperature or 57 °C (For more yield). If you want more concentration add less PE (50 µl).
13. Spin for 1 min at 13000 rpm to elute DNA from the column. Store DNA solution at -20 °C.

