

MEBEP TECH(HK) Co., Limited

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Plant Genomic DNA Fast Kit

Product Number: DNK3401

Shipping and Storage

- Buffer AP1 and AP2 may experience precipitation and precipitation at low temperatures. They can be re dissolved by taking a
 water bath at 37°C-65°C for a few minutes, without violent shaking to avoid excessive foam formation. After restoring clarity
 and transparency, cool to room temperature before use.
- 2. To avoid volatilization, oxidation, and pH changes caused by prolonged exposure of reagents to the air, each solution should be covered tightly in a timely manner after use.

Components

| Component | Storage | DNK3401 | DNK3402 | DNK3403 |
|------------------|---------|----------|-----------|-----------|
| | | 50 Preps | 100 Preps | 200 Preps |
| Buffer AP1 | RT | 20 ml | 40 ml | 80 ml |
| Buffer AP2 | RT | 7 ml | 13 ml | 26 ml |
| Buffer DD | RT | 10 ml | 10 ml | 20 ml |
| RNase A(10mg/ml) | 4°C | 250µl | 500µl | 1ml |

Description

This reagent kit adopts a unique buffer system, which is particularly suitable for extracting genomic DNA from plant dry powder or fresh plant materials. No need for phenol/chloroform extraction, safe and convenient to use, can maximize the removal of impurity proteins and other organic compounds in cells. There is no limit on the starting weight of the sample, and the experimenter can adjust it flexibly according to their own needs. The extracted genomic DNA fragments are large, highly purified, and of stable and reliable quality. The DNA recovered using this kit can be used for various routine operations, including enzyme digestion, PCR, library construction, Southern hybridization, and other experiments.

Features

- 1. No need to use toxic reagents such as phenol and chloroform.
- 2. Fast and simple, the entire operation process can be completed within 1 hour.
- 3. The results are stable and the yield is high (more than twice that of centrifugal column type), with a typical OD260/OD280 ratio of 1.7~1.9 and a length of 50kb-150kb. It can be directly used for PCR, Southern blot, various enzyme digestion reactions, and library construction.

Note

- 1. All centrifugation steps are completed at room temperature using a traditional desktop centrifuge with a speed of up to 13000 rpm.
- 2. Users need to bring their own isopropanol, 70% ethanol, liquid nitrogen mortar, and water bath.
- 3. Preheat the required water bath before starting the experiment for later use.
- 4. This reagent kit is a solution type and can easily increase or decrease the amount of each treatment in proportion. Please contact us to obtain operating manuals for other treatment amounts.

Protocol(Please read the precautions before the experiment)

- 1. Take an appropriate amount of plant tissue (100mg of fresh tissue or 20mg of dry weight tissue) and add liquid nitrogen to a mortar to grind thoroughly into fine powder.
- 2. Transfer the fine powder to a 1.5ml centrifuge tube, do not thaw, add 400ul Buffer AP1 and 4ul RNaseA (10mg/ml), vortex and

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shake well to aid in cracking.

If tissue lysis is difficult, a gentle 10 second step of homogenization can be added as needed to assist in lysis. In most cases, centrifugation is not necessary to remove partially lysed tissue, as there is a subsequent step of centrifugation removal.

- 3. 65°C water bath for 10 minutes, invert the centrifuge tube 2-3 times during the water bath process, and mix the sample.
- 4. Add 130ul Buffer AP2, mix well, let it stand on ice for 5 minutes, centrifuge at 14000rpm for 5 minutes, carefully aspirate the supernatant into a new 1.5ml centrifuge tube, being careful not to aspirate any interface substances.
- 5. **Optional steps:** Centrifuge the supernatant again at 14000 rpm (~13400×g) for 5 minutes, carefully and slowly aspirate the supernatant into a new 1.5ml centrifuge tube, do not aspirate the sediment.

The purpose of this step is to remove precipitated impurities from the supernatant, resulting in a higher purity of extracted genomic DNA.

- 6. Add 0.7 volume of room temperature isopropanol (for example, add 350ul of isopropanol to 500ul of supernatant), invert 30 times, mix well, or until a cotton like (filamentous) white DNA precipitate appears.
- 7. Centrifuge at 12000 rpm for 2 minutes, and white DNA precipitates can be seen at the bottom of the tube. Discard the supernatant.
- 8. Add 1ml of 70% ethanol, invert and rinse the DNA precipitate several times, centrifuge at 12000rpm for 1 minute, remove the supernatant (be careful not to pour out the DNA precipitate), invert and gently tap on absorbent paper a few times to control the residual ethanol. You can also use a gun to carefully suck out the residual ethanol around the bottom precipitate and the wall of the tube, and air dry the precipitate for a few minutes.

Be careful not to dry too much, otherwise DNA is extremely insoluble; Also, too much ethanol should not be left behind, otherwise ethanol may inhibit downstream reactions such as enzyme digestion.

- 9. Add an appropriate amount of Buffer DD to rehydrate and dissolve the DNA precipitate. Gently flick the tube wall and mix well, allowing it to incubate at 65°C for 30-60 minutes (no more than one hour). During this time, occasionally flick the tube wall to help rehydrate the DNA. DNA can also be rehydrated by staying overnight at room temperature or 4°C.
- 10. DNA can be stored at 2-8°C, and if stored for a long time, it can be stored at -20°C.