For research use only Version Number：2.0-1909

Plant DNA Isolation Kit

For genomic DNA purification from various plant tissue

Kit content	DE-06111	DE-06112	DE-06113
	50T	100T	250T
Buffer PL1	30 ml	60 ml	150 ml
Buffer PL2 *	30 ml	60 ml	150 ml
Buffer PW *	25 ml	50 ml	125 ml
Buffer WB	25 ml	50 ml	125 ml
Buffer EB	10 ml	20 ml	50 ml
Foregene Protease	1 ml	1 ml × 2	5 ml
DNA-Only Column	50	100	250
Manual	1	1	1

*： Buffer PL2 and Buffer PW contain irritating desalted salt. Please wear gloves and take relevant protective measures when operating.

Product introduction

This kit uses a DNA-only Column that can specifically bind DNA, a brand new Foregene Protease and a unique buffer system, which greatly simplifies the process of plant genomic DNA extraction and can obtain high-purity genomic DNA within 30 minutes, largely avoiding the degradation of genomic DNA.

The DNA-only silica gel membrane in the centrifugal column can efficiently and specifically adsorb DNA, without adding any organic reagent, it can remove RNA impurities, protein ions and organic compounds such as polysaccharide polyphenols to the maximum extent. The obtained DNA fragments are large, pure, high quality, stable and reliable.

Storage Condition

The kit can be stored for 12 months under dry conditions at room temperature (15–25℃). If you need to store longer time can be placed in 2-8℃. Foregene Protease solution has a unique formula, stored at room temperature for a long time (3 months) with activity; If stored at 4℃, its activity and stability will be better, so it is recommended to store at 4℃, remember not to store at-20℃.

Note: Solution is prone to precipitate if stored at low temperature. Before use, be sure to place the solution in the kit at room temperature for a period of time, if necessary, can be preheated in 37℃ water bath for 10 minutes, to dissolve precipitation, mixed before use.

DNA yield and Quality

The yield of DNA purified by Plant DNA Isolation Kit is related to the freshness of the source sample, preservation condition, water content and other factors. The following is the yield and purity of DNA extracted from each source sample using this Kit, which may be different from the data in actual operation.

Fresh leaves(100 mg)	DNA yield (μg)	OD260/280	OD260/230
Corn	4-7	1.7-1.9	1.8-2.1
Soybean	5-7	1.7-1.9	1.8-2.1
Wheat	10-14	1.7-1.9	1.8-2.1
Cotton	3-5	1.7-1.9	1.8-2.1
Rice	4-6	1.7-1.9	1.8-2.1
Tobacco	5-7	1.7-1.9	1.8-2.1
Oilseed rape	2-3	1.7-1.9	1.8-2.1

Note: The data in this table is for reference only. Due to the storage conditions of the materials used, operation proficiency and other factors, the data obtained may be slightly different from the data in this table.

Precautions:

- The sample should avoid repeated freezing and thawing, otherwise the extracted DNA fragment will be small and the extracted quantity will be reduced.
- Do not use more than 100 mg of fresh plant leaves or tissue and 30mg of dried plant tissue, otherwise DNA yield and purity will be affected.
- Before use, carefully check whether there is precipitation in Buffer PL1, Buffer PL2 and Buffer PW. If there is precipitation, please dissolve it at 37℃ and mix it before use.
- Before using the kit, be sure to check whether anhydrous ethanol is added to Buffer WB as instructed. Before Buffer WB was used, 60mL, 120 ml and 300 ml of anhydrous ethanol (DE-06111) were added, respectively.
- Elution volume: Buffer EB should not be less than 100μl, otherwise DNA production will be

affected.

- Do not add RNase to any Buffer.
- All centrifugation steps are performed in a table top centrifuge at room temperature (15-25℃).
- All experimental procedures were performed at room temperature (15-25℃).

Operating Steps

Before use, please add anhydrous ethanol to Buffer WB. Please refer to the label on the bottle for the added volume.

1. Add 600μl Buffer PL1 into a 2mL centrifuge tube, add 20μl Foregene Protease and 2μl β-mercaptoethanol (prepared by yourself), mix and put in a metal bath or water bath at 65℃ for preheating.
2. Take a proper amount of fresh plant leaves or tissue, dry tissue, as far as possible, cut into pieces, put in a pre-cooled mortar, add liquid nitrogen to fully grind.
3. Quickly weigh 100mg of ground fresh plant leaves or tissue powder or 30mg of dried tissue powder and transfer to Buffer PL1 preheated at 65℃. After the mixing is reversed quickly, the centrifugal tube is placed in a 65℃ metal bath or water bath for 10 min, and the mixing is reversed once every 5 min.

   Note: The sample powder is transferred immediately after grinding, otherwise the genomic DNA will degrade rapidly.
4. Add 600μl Buffer PL2, mix well, and put it back in a metal bath or water bath at 65℃ for 10 min.
5. 12,000 rpm (~13,400×g) centrifuge 5 min.
6. Transfer the supernatant to a new centrifugal tube using a micropipette.

   Note: try not to absorb the precipitate. If there are more solid impurities in the supernatant, repeat steps 5 and 6.
7. Add 180μl ethanol (96-100%), scroll thoroughly and mix. Flocculent precipitation may appear at this time.
8. Place the Column in a collection tube. Add 800 μl of mixed liquid to the DNA-Only Column, centrifuge at 12,000 RPM (~13,400×g) for 30Sec. Discard the waste liquid from the collection tube.
9. The centrifuge column was put back into the collection tube, and the remaining mixed liquid was added to the centrifuge column at 12,000 RPM (~13,400×g) for 1min. The waste liquid in the collection tube was discarded.
10. Add 500μl Buffer PW to the column, centrifuge at 12,000 RPM (~13,400×g) for 30Sec, discard the waste liquid from the collection tube.
11. Add 700μl Buffer WB to the centrifuge column (check whether absolute ethanol has been added before use), centrifuge at 12,000 RPM (~13,400 ×g) for 30 Sec, discard the waste liquid from the collection tube.
12. Repeat Step 11.
13. Put the column back into the collecting tube and centrifuge in empty tube at 12,000 RPM (~13,400×g) for 2 min.
14. Transfer the column to a new 1.5mL centrifuge tube, add 100μl of Buffer EB preheated at 65℃ to the center of the membrane (do not add the eluent to the pressure ring, otherwise a large volume of the eluent will be lost), and leave at room temperature for 2 min. Centrifugation at 12,000 RPM (~13,400×g) for 1 min. Add 100μl preheated Buffer EB to the center of the film, centrifuge at 12,000 RPM (~13,400×g) for 1min. Collect the eluent from the two collections.

    Note: If you wish to increase the concentration of DNA, the solution obtained from the first centrifugation can be added back to the column and centrifuged at 12,000 RPM (~13,400×g) for 1 min.