

## Plant DNA Preparation Kit

### DNA purification from plant tissue

#### Solution based genomic DNA Preparation Kit

	Cat.-No.	Amount
	PP-207XS	20 preparations
	PP-207S	100 preparations
	PP-207L	500 preparations

For *in vitro* use only.

Quality guaranteed for 12 months.

Store at room temperature.

For long term storage place *RNase A lyophilisate* at -20°C.

*RNase A Solutions* should be stored at -20°C.

#### Kit contents

Cell Lysis Solution 1%

*RNase A* (before use, solve in double distilled water to obtain a final concentration of 4 mg/ml)

Protein Precipitation Solution

Washing Buffer (before use, add 96-99% Ethanol as indicated on the bottle)

DNA Hydration Solution

#### To be provided by you

Isopropanol (2-propanol) >99%

96-99% Ethanol

Microtubes 1.5 ml

#### Description

Plant DNA Preparation Kit is designed for convenient and fast isolation of genomic DNA from plant tissue. The solution based system minimizes DNA fragmentation that may be problematic in other spin-column/filtration based method. Because phenol or chloroform is not used it is safe and does not produce any harmful waste.

Solution based genomic DNA purification kits guarantee minimal DNA fragmentation and yield DNA sized up to 150 kb.

#### Expected yield

Yields of genomic DNA will vary from sample to sample depending on the amount, quality and type of material processed. An amount of approx. 0.25 µg purified DNA per preparation can be expected.

Buffer	PP-207XS	PP-207S	PP-207L
	20 preps	100 preps	500 preps
Cell Lysis Solution 1%	6.4 ml	32 ml	160 ml
<i>RNase A</i>	0,16 mg (before use, solve in 40 µl dd-water to obtain a final concentration of 4 mg/ml)	0.8 mg (before use, solve in 200 µl dd-water to obtain a final concentration of 4 mg/ml)	5 x 0.8 mg (before use, solve each vial in 200 µl dd-water to obtain a final concentration of 4 mg/ml)
Protein Precipitation Solution	2.2 ml	11 ml	55 ml
Washing Buffer	add 9.6 ml Ethanol (final volume 12 ml)	add 48 ml Ethanol (final volume 60 ml)	add 120 ml Ethanol to each bottle (final volume 150 ml each)
DNA Hydration Solution	2.2 ml	11 ml	55 ml

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#### Preparation procedure

Before start, provide >99% Isopropanol (2-propanol) (not included in the kit).

**For XS pack (20 preps):** Add 40 µl dd-water to the RNase A tube and 9.6 ml 96-99% Ethanol (not included in the kit) to the Washing Buffer bottle.

**For S pack (100 preps):** Add 200 µl dd-water to the RNase A tube and 48 ml 96-99% Ethanol (not included in the kit) to the Washing Buffer bottle.

**For L pack (500 preps):** Add 200 µl dd-water to each RNase A tube and 120 ml 96-99% Ethanol (not included in the kit) to each Washing Buffer bottle.

The RNase A Solution should be stored at -20°C.

#### 1. Sample collection and Handling

- Fresh or frozen tissue may be finely ground with a mortar and pestle in liquid nitrogen prior to DNA isolation.
- Work quickly and keep tissue cold to minimize DNase activity.

#### 2. Cell Lysis

- Transfer the finely ground tissue (10-30 mg) to a 1.5 ml microtube.
- Add 300 µl Cell Lysis Solution to the tissue.
- Incubate at 65°C for 60 min.
- Invert the tube occasionally during the incubation.

#### 3. RNase Treatment

- Add 1.5 µl of RNase A Solution to the cell lysate.
- Mix the sample by inverting the tube 25 times and incubate at 37°C for 15-60 min.

#### 4. Protein Precipitation

- Cool the sample to room temperature and add 100 µl of Protein Precipitation Solution to the cell lysate.
- Mix the solution well by vortexing.
- Centrifuge at 15,000 g for 3 min. (The precipitant should form a tight, green pellet. If the pellet is not tight, repeat mixing, incubate on ice for 10 minutes, and then centrifuge again.)

#### 5. DNA Precipitation

- Transfer the DNA containing supernatant to a clean 1.5 ml microtube containing 300 µl of Isopropanol >99%.
- Mix the sample by inverting gently 50 times.
- Centrifuge at 15,000 g for 1 min. The DNA will be visible as a pellet that ranges in color from off-white to light green.
- Discard the supernatant and drain tube briefly on clean absorbent paper.
- Add 500 µl Washing Buffer and invert tube several times to wash the DNA Pellet.
- Centrifuge at 15,000 g for 1 min.
- Discard the ethanol carefully.
- Air dry at room temperature for 10-15 min.

#### 6. DNA Hydration

- Add 50-100 µl of DNA Hydration Solution to the dried DNA pellet.
- Hydrate the DNA by incubating sample at 65°C for 60 min.
- Store DNA at 4°C. For long time storage, place sample at -20°C or -80°C.