Pierce™ Plant Total Protein Extraction Kit

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WARNING! Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves. Safety Data Sheets (SDSs) are available from **thermofisher.com/support**.

Product description

The Thermo Scientific™ Pierce™ Plant Total Protein Extraction Kit is composed of optimized buffers and filter cartridges that allow for efficient and rapid protein extraction in less than 10 minutes. The kit is designed to rapidly extract denatured or native proteins from plant tissues (leaves, seeds, soft stem and roots etc.). This kit provides both denaturing and native cell lysis buffers to allow flexibility based on the desired downstream application. The protein extracts can be used for applications such as SDS-PAGE, Western blotting, immunoprecipitation, affinity purification and activity assays.

Contents and storage

Product[1]	Cat. No.	Contents[2]	Amount	Storage
Pierce™ Plant Total Protein Extraction Kit	A44056	Denaturing Lysis Buffer	25 mL	Room temperature
		Native Lysis Buffer	25 mL	
		Protein Extraction Filter Cartridges	50 each	
		Collection tubes	50 each	
		Plastic rods	2 each	

[1] 50 preparations of 50–200mg of plant tissue each.
[2] Components are supplied by Invent Biotechnologies, Inc., Plymouth, MN.

Procedural guidelines

- Protease and/or phosphatase inhibitors (Thermo Scientific™ Halt Protease Inhibitor Cocktail or Thermo Scientific™ Halt Phosphatase Inhibitor Cocktail) are required to maintain extract integrity and function. Immediately before use, add inhibitors to the volume of lysis buffer required and keep tissue, buffers, and extracts on ice.
- Typical protein yield is 1–6 mg/mL, however, it is possible to end up with highly concentrated protein extracts, especially from leaves (up to 20 mg/mL). Dilution may be required for protein estimation, downstream applications, and long term storage at –20°C. The addition of glycerol to 10–20% is also suggested for long-term storage at –20°C.
- The extracts are compatible with the Pierce™ BCA Protein Assay and the Pierce™ Rapid Gold BCA Protein Assay.
- If precipitate is seen in the Denaturing Lysis Buffer, incubate at 37°C to completely dissolve.
- For dry seeds, soak the seeds in water for two days before use.
- The plastic rod is reusable; clean and rinse it thoroughly with distilled water.
- Each filter cartridge can process 50 to 200 mg of tissue.

Use Denaturing Lysis Buffer to prepare tissue

1. Prepare the tissue (per 100 mg) for extraction:
   - **For plant leaves**, place fresh tissue in the filter cartridge by folding the leaves into a smaller size. Punch the leaf in the filter repeatedly with a 200/1000 µL pipette tip approximately 60 times.
   - **For seeds** and **soft stems** cut into smaller pieces with a sharp blade or scissors and place into the filter cartridge; grind with plastic rod approximately 60 times with twisting force.

2. Add 100 µL Denaturing Lysis Buffer to the filter.
3. Grind the tissue with the plastic rod approximately 60 times with twisting force.
4. Cap the filter and incubate at room temperature for 1–2 minutes.
5. Centrifuge at 16,000 × *g* for 5 minutes.
6. Transfer supernatant (the denatured total protein extract) to a new tube.

Use Native Lysis Buffer to prepare tissue

1. Prepare the tissue (per 100 mg) for extraction:
   - **For plant leaves**, place fresh tissue in the filter by folding the leaves into a smaller size and place into the filter cartridge. Punch the leaf in the filter repeatedly with a 200/1000 µL pipette tip approximately 60 times.
   - **For seeds** and **soft stems** cut them into smaller pieces with a sharp blade or scissors and place into the filter cartridge; grind it with plastic rod approximately 60 times with twisting force.
2. Add 100 µL Native Lysis Buffer to the filter.
3. Grind the tissue with the plastic rod approximately 60 times with twisting force.
4. Cap the filter and incubate on ice for 5 minutes.
5. Centrifuge at 16,000 × *g* for 5 minutes.
6. Transfer supernatant (the native total protein extract) to a new tube.

Troubleshooting

Observation	Possible cause	Recommended action
Low protein activity	Extract is degraded.	Keep sample cold and use protease and/or phosphatase inhibitors.
Low protein concentration	Not enough lysis buffer for the amount of tissue used.	Increase the lysis buffer volume by 50 µL.
	Not enough starting tissue used.	Use up to 200 mg of tissue.
	Tissue pieces are too large for extraction.	Ensure pieces are small and well ground with the plastic rod.

Limited product warranty

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Thermo Fisher Scientific | 3747 N. Meridian Road | Rockford, Illinois 61101 USA
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