# Minute<sup>TM</sup> Detergent-Free Plant Protein Extraction Kit

#### Catalog Number: SN-010

# Description

Invent Biotechnologies Minute<sup>TM</sup> detergent-free plant protein extraction kit is composed of optimized protein extraction buffer and protein extraction filter cartridges with 2.0 ml collection tubes. The kit is designed to rapidly extract water soluble proteins from plant tissues. The protein extraction buffer does not contain any detergent and organic solvent. Due to the use of the protein extraction filter cartridges, the extraction volume can be as small as 50 µl and as large as 500 µl. Detergent-free total proteins can be extracted from plant tissues (leaves, seeds, soft stems etc.) in less than 8 min with high yield (1-5 mg/ml).

## Application

Minute<sup>™</sup> detergent-free plant protein extraction kit is designed to rapidly extract water soluble proteins from plant tissues for applications such as proteomics (LC/MS), IP, ELISA, 2D-gel analysis, isoelectric focusing, SDS-PAGE, immunoblottings, and other applications. This kit provides a very rapid method for preparation of high concentration of protein extract

## **Buffer Formulation:** Proprietary

## **Kit components**

- 1. 25 ml DF-lysis buffer
- 2. 50 protein extraction filter cartridges
- 3. 50 collection tubes with cap
- 4. Plastic rods (4)

**Storage:** Store the kit at 4°C

## **Important Product Information**

The Minute<sup>TM</sup> detergent-free protein extraction kit is designed to extract water soluble proteins rapidly. The use of protease inhibitors is not necessary prior to extraction. However if downstream application takes significant amounts of time or the protein extract will be stored for longer period of time, addition of protease inhibitors to protein extraction buffer is recommended. For determination of protein concentration, BCA kit (Pierce) is recommended. To study protein phosphorylation, **phosphatase inhibitors** (such as PhosStop from Roche) should be added to the extraction buffer prior to use.

#### **Additional Materials Required**

Table-Top Microcentrifuge BCA Protein Assay Kit (Pierce, Cat #. 23227)

#### **Protein Extraction Procedures**

Following procedures are for 50-100 mg starting plant tissues (fresh leaves, seeds and soft stem and roots etc.). For dry seeds, soak in water for two days. If smaller or larger amounts of starting materials are used, adjust the amount of protein extraction buffer proportionately.

- 1. Prior to protein extraction pre-chill protein extraction buffer and the protein extraction filter cartridge in collection tube on ice.
- 2. For plant leaves, place 50-100 mg fresh tissue in the filter by folding or rolling the leaves into smaller volume and insert into the filter cartridge. Punch the leaf in the filter repeatedly with a pipette tip for 50-60 times and go to step 3 (for tissues less than 50 mg punching is not necessary). For seeds (fresh/frozen) and soft stems cut them into smaller pieces with a sharp blade and place them in the filter cartridge; grind it with plastic rod with twisting force for 50-60 times and go to step 3.
- 3. Add 50-100  $\mu$ l DF-lysis buffer to the filter (note: shake the bottle vigorously to resuspend tissue dissociation beads prior to use). Grind the tissue with a plastic rod (included) for 50-60 times with twisting force (Note: The plastic rod is reusable, for cleaning, rinse it thoroughly with distilled water and dry it with paper towel).
- 4. Cap the filter and incubate on ice for 5 min. Centrifuge using a microcentrifuge at top speed for 2-5 min. Transfer supernatant to a fresh tube (this is detergent-free protein extract).

Important Note: the presence of some un-lysed tissue would not affect the quality of the samples.