

## Detergent-Free Total Protein Isolation Kit

## Product Insert

Product #: 30300

Norgen's Detergent-Free Total Protein Isolation Kit provides a fast and simple procedure for the isolation of total proteins from tissue, bacteria, yeast or mammalian cells without the use of SDS, Triton® X-100 and other detergents. Detergents are extensively used to prepare protein samples; however, these detergents have undesirable effects on downstream analysis. These effects include extraneous peaks in mass spectrometry, artifacts with chromatography and electrophoresis, interference with microinjection into cells and interference with protein immunization.

The Detergent-Free Total Protein Isolation Kit maintains high protein recovery. Purification is based on using Norgen's proprietary resin together with Lysis Solution, followed by protein filtration using Norgen's filter column. The purified proteins can be used in a number of downstream applications including mass spectrometry, SDS-PAGE, isoelectric focusing, and NMR spectroscopy.

The Detergent-Free Total Protein Isolation Kit contains all the solutions, tubes and columns for the processing of 25 total detergent-free protein samples. Each Lysis Tube is able to process up to 50 mg of tissue,  $10^{10}$  bacterial cells,  $10^9$  yeast cells or  $10^7$  mammalian cells. Preparation time for 12 samples is less than 10 minutes. The kit has a shelf life of at least 1 year when stored as suggested.

### Kit Specifications

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Maximum Amount of Starting Material:	
Tissues	50 mg
Animal Cells	$1 \times 10^7$ cells
Yeast	$1 \times 10^9$ cells
Bacteria	$1 \times 10^{10}$ cells
Time to Process 12 Samples	Less than 10 minutes

### Kit Components

Component	Product # 30300 (25 samples)
Lysis Solution	4 mL
Lysis Tubes	25
Filter Columns	25
Elution tubes (1.7 mL)	25
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### **Storage Conditions and Product Stability**

The Lysis Solution should be kept tightly sealed and stored at room temperature. Once opened, the solution should be stored at 4°C. This kit is stable for 2 years after the date of shipment.

### **Precautions and Disclaimers**

This kit is designed for research purposes only. It is not intended for human or diagnostic use.

Ensure that a suitable lab coat, disposable gloves and protective goggles are worn when working with chemicals. For more information, please consult the appropriate Material Safety Data Sheets (MSDSs). These are available as convenient PDF files online at [www.norgenbiotek.com](http://www.norgenbiotek.com).

### **Customer-Supplied Reagents and Equipment**

- Benchtop microcentrifuge
- Micropipettors
- Vortex

## **Procedure**

### **Notes prior to use**

- Briefly spin the Lysis Tubes prior to use in order to collect the grey material into the bottom of the tube
- Maximum input amount for each Lysis Tube is up to 50 mg of tissue,  $10^{10}$  bacterial cells,  $10^9$  yeast cells or  $10^7$  mammalian cells

### **1A. Sample Lysis - Tissue**

- a. Obtain the desired tissue sample.
- b. Add the tissue sample to the **Lysis Tube**.
- c. Add 50  $\mu$ L of the **Lysis Solution** to the **Lysis Tube** containing the tissue sample.
- d. Cap the **Lysis Tube** and vortex vigorously for 2 minutes.
- e. Centrifuge the **Lysis Tube** at 14,000 x g for 2 minutes.

### **1B. Sample Lysis – Bacteria, Yeast or Mammalian Cells**

- a. Obtain the sample, and pellet the cells at the appropriate centrifugation speed (5 minutes at 2,000 rpm for mammalian cells or 1-2 minutes at 14,000 for yeast and bacterial cells).
- b. Completely discard the supernatant and resuspend the cells in 50  $\mu$ L of the **Lysis Solution**.
- c. Add the resuspended cells to the **Lysis Tube**.
- d. Cap the **Lysis Tube** and vortex vigorously for 2 minutes.
- e. Centrifuge the **Lysis Tube** at 14,000 x g for 2 minutes.

### **2. Sample Filtration**

- a. Place a **Filter Column** into a provided **Elution Tube**.
- b. Transfer the liquid content from the **Lysis Tube** into the **Filter Column**.
- c. Centrifuge at 14,000 x g for 1 minute, ensure that the entire sample has passed through into the Elution Tube.

### **3. Sample Storage**

Flash freezing at -20°C is recommended to avoid protein degradation. In addition, the desired protease inhibitor should be added to the isolated proteins, mixed well and stored at -20°C. It is recommended that the protease inhibitor used does not interfere with any downstream application or assays

**Total proteins are now ready for downstream applications.**

<b>Related Products</b>	<b>Product #</b>
ProteoSpin™ Detergent Clean Up Micro Kit	10200, 10500
ProteoSpin™ Detergent Clean Up Maxi Kit	17100

**Technical Support**

Contact our Technical Support Team between the hours of 8:30 and 5:30 (Eastern Standard Time) at (905) 227-8848 or Toll Free at 1-866-667-4362.

Technical support can also be obtained from our website ([www.norgenbiotek.com](http://www.norgenbiotek.com)) or through email at [techsupport@norgenbiotek.com](mailto:techsupport@norgenbiotek.com).

Norgen's purification technology is patented and/or patent pending. See [www.norgenbiotek.com/patents](http://www.norgenbiotek.com/patents)

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