

## Inclusion Body Solubilization Reagent

Product # 18700 (25 mL)

Product # 18701 (100 mL)

## Product Insert

Overexpression of recombinant proteins in bacteria frequently results in the formation of insoluble aggregates termed inclusion bodies. Solubilization of inclusion bodies is a necessary step to release the recombinant protein, and is traditionally accomplished through denaturation with chaotropic reagents. These reagents disrupt hydrogen bonds and hydrophobic interactions so that the protein can be further purified by chromatographic means. More importantly, denaturation of the incorrectly folded protein is often a step that is taken to initiate a refolding process so that the protein can achieve its native conformation for full biological activity.

Norgen's Inclusion Body Solubilization Reagent dissolves inclusion body aggregates resulting from the expression of recombinant proteins in bacteria. Once dissolved, proteins can be analyzed by SDS-PAGE, quantified or further purified for refolding.

### Product Components

| Component                             | Product # 18700 | Product # 18701 |
|---------------------------------------|-----------------|-----------------|
| Inclusion Body Solubilization Reagent | 25 mL           | 100 mL          |
| Product Insert                        | 1               | 1               |

### Storage Conditions and Product Stability

The Inclusion Body Solubilization Reagent should be stored at 4°C. This reagent should remain stable for at least 1 year in its unopened container.

### Precautions and Disclaimers

This reagent 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

- Centrifuge
- Appropriate centrifuge tubes
- Vortex

## Procedure

### Notes prior to use:

- Ensure that no precipitates have formed. If necessary, warm and mix well until the solutions become clear again.

## Procedures

### A. Small Scale Extraction (from 1.5 mL of induced culture)

1. Purify inclusion body protein pellet using Norgen's **Cell Lysis Reagent** (Product No. 18800/18801) or another method.
2. Add 50  $\mu$ L of **Inclusion Body Solubilization Reagent** to pellet. To increase or decrease protein concentration, adjust the amount of reagent used accordingly.
3. Resuspend pellet using pipette and vortex until the pellet is fully dissolved.
4. Remove cell debris by centrifugation at 14,000 rpm for 1 minute.
5. Transfer supernatant (containing solubilized protein) to a fresh tube for further analysis.

### B. Large Scale Extraction (from 50 to 500 mL of induced culture)

1. Purify inclusion body protein pellet using Norgen's **Cell Lysis Reagent** (Product No. 18800/18801) or another method.
2. To maximize protein concentration, it is recommended that the inclusion body pellet is dissolved in a minimal amount of reagent. Begin by adding 1 mL of **Inclusion Body Solubilization Reagent** to pellet.
3. Resuspend pellet using pipette and vortex until the pellet is fully dissolved.
4. If the pellet is not completely dissolved after ample pipetting and vortexing, add additional **Inclusion Body Solubilization Reagent** in 1 mL increments, or incubate on a rocker at room temperature to aid solubilization.
5. Remove cell debris by centrifugation at 14,000 rpm for 10 minutes.
6. Transfer supernatant (containing solubilized protein) to a fresh tube for further analysis.

| <b>Related Products</b>  | <b>Product #</b> |
|--------------------------|------------------|
| Cell Lysis Reagent       | 18800            |
| Inclusion Body Micro Kit | 10300            |
| Inclusion Body Maxi Kit  | 17700            |
| ProteoLadder 100         | 12300            |

### **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).

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