G-Biosciences ♦ 1-800-628-7730 ♦ 1-314-991-6034 ♦ technical@GBiosciences.com

A Geno Technology, Inc. (USA) brand name

Gram-Negative Lysis Buffer

Lysis buffer for Gram-negative bacteria

(Cat. # 786-566, 786-567, 786-579, 786-580)

INTRODUCTION

Gram-negative Lysis Buffer is an extraction buffer for soluble proteins from Gram-negative bacteria. It is a proprietary improvement on the lysozyme based lysis in combination with various salts and agents, which allows extraction of soluble proteins. Depending on the application, additional agents such as reducing agents, chelating agent, nuclease and protease inhibitors cocktail may be added into Gram-negative lysis buffer *(see Related Products for protease inhibitor Protease Arrest™).* This buffer has been tested for use with several widely used bacteria including *E. coli* strains.

ITEMS SUPPLIED

Cat. #	Description	Size
786-566	Gram-Negative Lysis Buffer	125ml
786-567	Gram-Negative Lysis Buffer	250ml
786-579	Gram-Negative Lysis Buffer	500ml
786-580	Gram-Negative Lysis Buffer	1L

STORAGE CONDITIONS

It is shipped at ambient temperature. Upon arrival, store at room temperature and is stable for 1 year.

ADDITIONAL ITEMS NEEDED BUT NOT SUPPLIED

Lysozyme, Nuclease cocktail (DNase & RNase), Protease Inhibitors cocktail (see related products), Centrifuge, Test tubes

INSTRUCTIONS FOR USE

1. Pellet the bacterial cells by centrifuging the liquid culture at 5,000 × g for 10 minutes. Remove the supernatant and re-suspend the wet bacterial cell pellet in 1x PBS *(500ul PBS/50ul wet pellet)* by gently flicking the bottom of the tube.
2. Add lysozyme 5ul/50ul wet pellet *(see related product Longlife Lysozyme)* into the bacterial suspension. Mix it gently and incubate the suspension at 37^0C for 60 minutes.
3. After incubation, centrifuge the suspension at 5,000 x g for 10 minutes. Remove the supernatant carefully without disturbing the spheroplast pellet.

4. Add 1 ml of Gram-Negative Lysis Buffer per 50ul of wet bacterial cell pellet. **Optional:** Add protease inhibitors for inhibiting protease activity *(see Related Products Bacterial Protease Arrest)* and/or nuclease cocktail *(see Related Products Longlife Nuclease)* for removal of nucleic acids *(DNA & RNA)* released during the cell lysis *(usually 5ul/50ul wet bacterial pellet)*.
5. Vortex the tube and pipette the lysate up and down to get a clear lysate containing proteins.
6. Centrifuge the lysate at 20,000 x g for 15 minute and collect the clear lysate from residual cell debris/insoluble proteins.

RELATED PRODUCTS

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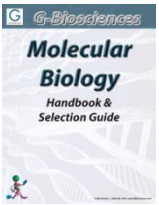

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