

Product Information

LMNG (Lauryl Maltose Neopentyl Glycol)

Catalog Numbers: LMN01, LMN02, LMN03

Description

LMNG (Lauryl Maltose Neopentyl Glycol) is a new-generation MNG-class amphiphilic detergent designed for high-performance membrane-protein research. Compared with conventional detergents, LMNG features a unique neopentyl-glycol core scaffold (double-head, double-tail structure), delivering mild yet efficient solubilization capacity. It preserves the native conformation and functional activity of membrane proteins to the greatest extent during extraction.

LMNG can be used in combination with other detergents. It is applicable for extraction and purification of unstable membrane proteins, such as GPCRs and ion channels, sample preparation for structural biology, including cryo-EM and protein crystallization, as well as studies on drug-target stability and biomolecular interaction analysis.

Applications

- Extraction and purification of unstable membrane proteins, including GPCRs and ion channels
- Sample preparation for structural biology, such as cryo-EM and protein crystallization
- Drug-target stability studies
- Biomolecular interaction analysis

Transportation and Storage

- Ship at ambient temperature.
- Powder: -20 °C for 3 years
- In solvent: -80 °C for 1 year

Instructions for Use

Preparation of 10% (w/v) LMNG Stock Solution

Given the high hygroscopicity of LMNG, it is recommended to prepare a 10% (100 mg/mL) high-concentration stock solution for storage.

- 1) Weighing: Quickly weigh solid LMNG under well-ventilated and dry conditions.
- 2) Dissolution: Add deionized water (or purification buffer). Rotate end-over-end in a cold room, or shake vigorously; avoid prolonged sonication (foaming and local heating may degrade the detergent) until the solution becomes completely clear.
- 3) Filtration and aliquoting: Sterilize by filtration through a 0.22 µm membrane filter. Aliquot into single-use portions (e.g., 100 µL per tube). Store at -20 °C, protected from light. The stock solution is stable for more than 6 months; avoid repeated freeze-thaw cycles.

Experimental Procedure

This section uses human neurotensin receptor 1 (hNTSR1) and adenosine A2A receptor (A2AR) as examples to provide reference methods for membrane solubilization, extraction, and cobalt-ion affinity purification of GPCRs expressed by baculovirus using the LMNG-CHS system.

1. Materials

- 1) Detergents: Self-developed LMNG (Distolife, Cat:LMN01/LMN02/LMN03), self-developed CHS (Distolife, Cat: CHS01/CHS02/CHS03).
 - 2) Auxiliary reagents: Tris-HCl, HEPES, NaCl, CaCl₂, MgCl₂, glycerol, imidazole, protease inhibitor cocktail.
 - 3) Key consumables and equipment: 0.22 µm filter membrane, immobilized cobalt ion IMAC affinity chromatography resin, size exclusion SEC 250 analytical chromatography column, 4-12% SDS-PAGE gel, modular high-performance HPLC system, high-speed refrigerated centrifuge.
- (Please contact us if you need detailed information.)

2. Preparation of Detergent Solutions

- 1) 3 g of LMNG powder was dissolved in approximately 40 mL of 200 mM Tris-HCl (pH 8.0), and the solution was adjusted to a final volume of 50 mL to obtain a 6% (w/v) stock solution.

2) CHS was added to a final concentration of 1.2% (w/v), followed by sonication until the solution became completely clear and colorless. The solution was brought to a final volume of 50 mL. After sterile filtration through a 0.22 μ m filter membrane, the solution was aliquoted and stored at -80°C for subsequent use.

3. Expression of GPCR Membrane Proteins and Membrane Preparation

1) The Bac-to-Bac baculovirus expression system was used to construct recombinant baculoviruses encoding human GPCR family membrane proteins (including hNTR1 and A2AR). 50 mL ExpiSf9 insect cells were infected with the recombinant viruses for target protein expression.

2) Cell pellets were collected, thawed on ice, and resuspended in low-salt buffer (20 mM HEPES pH 7.4, 100 mM NaCl, 5 mM CaCl_2 , 5 mM MgCl_2 , 10% glycerol, supplemented with protease inhibitor cocktail).

3) The mixture was centrifuged at $100,000 \times g$ for 30 min at 4°C (e.g., 35,000 rpm in a Type 70 Ti rotor), and the cell membrane pellet was collected for downstream experiments.

4. Extraction and Purification of Membrane Proteins

1) Membrane resuspension and homogenization: The cell membrane pellet was resuspended in buffer (20 mM HEPES pH 7.4, 100 mM NaCl, 5 mM MgCl_2 , 5 mM CaCl_2 , supplemented with protease inhibitor cocktail). The suspension was fully homogenized using a Dounce homogenizer and divided equally into two aliquots of 1 mL each.

2) Membrane protein solubilization: LMNG & CHS working solution was added to the sample to final concentrations of 1% LMNG / 0.1% CHS (W/V) (or the final concentrations actually used). The mixture was incubated by rotation at 4°C for 3 h to achieve solubilization of membrane proteins.

3) Clarification and affinity binding: The sample was centrifuged at 35,000 rpm for 30 min, and the supernatant was harvested. 60 μ L of immobilized cobalt ion IMAC affinity chromatography resin (1 CV = 30 μ L) together with a solution containing 20 mM imidazole was incubated with the supernatant overnight at 4°C to allow binding of target proteins to the resin.

4) Washing and elution:

5) Washing step: Non-specifically bound impurities were removed by gradient washing with 20 column volumes (CV) of wash buffer (20 mM HEPES pH 7.4, 150 mM NaCl, 30/50 mM imidazole, 0.05% LMNG, 0.01% CHS).

6) Elution step: GPCR target proteins were eluted using 5 CV of elution buffer (20 mM HEPES pH 7.4, 150 mM NaCl, 250 mM imidazole, 0.01% LMNG, 0.002% CHS).

5. Sample Analysis

1) HPLC-SEC analysis: 60 μ L protein sample was loaded onto a Sepax Nanofilm SEC-250 column. Chromatograms were recorded to evaluate protein monodispersity and aggregation status.

2) SDS-PAGE analysis: 30 μ L of eluted protein sample was mixed thoroughly with 10 μ L of 4 $\times$ protein loading buffer. A total of 30 μ L of the mixture was loaded onto a 4–12% gradient SDS-PAGE gel for electrophoresis to assess the purity of the target proteins.

Notice

1. To ensure the safety and health of operators, laboratory coats and disposable gloves must be worn throughout the entire experimental procedure.
2. Certain technical information provided with this product is intended for customer reference, academic communication, and scientific research purposes only. Our company does not warrant the absolute authority or completeness of the information provided.
3. This product is for research use only. It is strictly prohibited for use in clinical diagnosis, treatment, food, pharmaceuticals, or any other applications involving human use. Do not store this product in ordinary residential premises.
4. The final LMNG concentration in the experimental system must be above its critical micelle concentration (CMC). Insufficient concentration will cause micelle dissociation and membrane protein precipitation.
5. When concentrating membrane proteins using ultrafiltration concentrators (e.g., 100 kDa MWCO), LMNG micelle aggregates have large particle sizes, so the detergent will be concentrated alongside the target protein. It is recommended to keep the LMNG concentration in the

eluate between 0.005% and 0.01%. Excessively high detergent concentration after concentration will interfere with downstream cryo-EM sample preparation (resulting in elevated background and thicker ice layers) or protein crystallization experiments.

Troubleshooting

Possible Problems and Solutions:

Symptoms	Solution
The solution does not become clear after dissolving the product in water	Dissolve the product in PBS buffer instead.
Some GPCRs exhibit high conformational heterogeneity and insufficient stability when solubilized with LMNG alone, which cannot meet the requirements for structural biology studies.	Combining CHS with LMNG can greatly improve the stability of GPCRs. CHS modifies detergent micelles to more closely mimic the native lipid bilayer. In practice, a weight ratio of LMNG: CHS = 10:1 is commonly used for mixed solubilization.