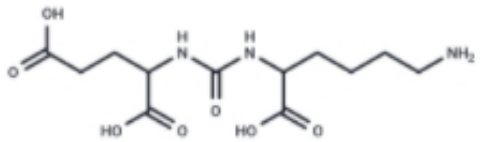
Product Data Sheet

Product Name: Glu-urea-Lys
Cat. No.: GC26528

Chemical Properties

Cas. No.	1025796-69-3		
Formula	$C_{12}H_{21}N_3O$	M.Wt	319.31
Solubility	DMSO: 50 mg/mL (156.59 mM)	Storage	-20°C,away from moisture
General tips	For obtaining a higher solubility , please warm the tube at 37 °C and shake it in the ultrasonic bath for a while.Stock solution can be stored below -20°C for several months.		
Shipping Condition	Evaluation sample solution : ship with blue ice All other available size: ship with RT , or blue ice upon request.		
Structure			



Protocol

This protocol is for reference only. The actual application conditions may need to be optimized according to your specific experimental needs.

Preparation of Glu-urea-Lys ligand-coupled lipid nanoparticle/siRNA system [1-2]:

1.Reagent preparation: DSPE-PEG2000-NH_2, N-hydroxysuccinimide (NHS), 1-ethyl-3-(3-dimethylaminopropyl) carbodiimide (EDC), borate buffer (0.1M, pH 8.5), dialysis bag (MWCO 3.5kDa), siRNA, citric acid buffer (10mM, pH 4.0),

2. Preparation of Glu-urea-Lys-DSPE-PEG2000 conjugate

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2.1 Carboxyl activation: Dissolve Glu-urea-Lys (1eq), NHS (2eq), and EDC (2eq) in DMF and stir at room temperature for 4h.

2.2 DSPE-PEG2000 coupling: The activated product was added dropwise to DSPE-PEG2000-NH_2 (1.2eq) in borate buffer, pH 8.5, and reacted at room temperature in the dark for 24h.

2.3 Purification: The reaction mixture was placed in a dialysis bag, sealed and immersed in PBS buffer (4°C, in the dark, 48h) to remove small molecule impurities, and the conjugate was obtained after freeze-drying.

2.4 Verification: MALDI-TOF MS was used to verify the molecular weight of the compound (expected molecular weight: DSPE-PEG2000 + Glu-urea-Lys ≈ 2800Da + 348Da).

3. Preparation of lipid nanoparticles (LNP) and siRNA loading

3.1 Preparation of lipid solution: Each lipid was dissolved in ethanol (total lipid concentration 10mg/mL) according to the ratio of DLin-MC3-DMA (50%), cholesterol (38.5%), DSPC (10%), and Glu-urea-Lys-DSPE-PEG (1.5%), and ultrasonicated in a 60°C water bath until completely dissolved.

3.2 Preparation of aqueous phase: siRNA was dissolved in citric acid buffer (0.2mg/mL).

3.3 Microfluidic mixing: The lipid phase and the aqueous phase were mixed at a flow rate ratio of 3:1 (total flow rate 12mL/min), and the outlet was collected in PBS (pH 7.4).

3.4 Dialysis purification: The reaction mixture was placed in a dialysis bag, sealed and immersed in PBS buffer (4°C, protected from light, 48h), and the buffer was changed 3 times during the period.

4. Quality control

4.1 Particle size: DLS detection (80-100nm, PDI < 0.2).

4.2 Encapsulation efficiency: RiboGreen method (> 95%).

4.3 siRNA integrity: agarose gel electrophoresis (no free siRNA bands).

Notes

1. Aseptic operation: In vivo experiments require filtration sterilization (0.22μm filter membrane).

Caution: Product has not been fully validated for medical applications. For research use only.
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2. siRNA stability: LNPs must be stored at -80°C after preparation to avoid repeated freezing and thawing.

3. In vivo distribution: Bioluminescence imaging (such as DiR-labeled LNPs) can be supplemented to observe targeting.

References:
[1]. Lee J B, Zhang K, Tam Y Y C, et al. A Glu-urea-Lys ligand-conjugated lipid nanoparticle/siRNA system inhibits androgen receptor expression in vivo[J]. Molecular Therapy Nucleic Acids, 2016, 5.
[2]. Zhou W, Huang J, Xiao Q, et al. Glu-Urea-Lys Scaffold Functionalized Superparamagnetic Iron Oxide Nanoparticles Targeting PSMA for In Vivo Molecular MRI of Prostate Cancer[J]. Pharmaceutics, 2022, 14(10): 2051.

Background

Glu-urea-Lys is a small molecule ligand that targets prostate-specific membrane antigen (PSMA). Glu-urea-Lys is composed of glutamic acid (Glu), urea bond (urea), and lysine (Lys), and can specifically bind to prostate cancer cells that highly express PSMA. Glu-urea-Lys can enhance its accumulation at the tumor site after being combined with liposomes, and is often used in the diagnosis or treatment of prostate cancer [1-4].

References:
[1]. Wüstemann T, Bauder-Wüst U, Schäfer M, et al. Design of internalizing PSMA-specific Glu-ureido-based radiotherapeuticals[J]. Theranostics, 2016, 6(8): 1085.
[2]. Baranski A C, Schäfer M, Bauder-Wüst U, et al. PSMA-11–derived dual-labeled PSMA inhibitors for preoperative PET imaging and precise fluorescence-guided surgery of prostate cancer[J]. Journal of Nuclear Medicine, 2018, 59(4): 639-645.
[3]. Lee J B, Zhang K, Tam Y Y C, et al. A Glu-urea-Lys ligand-conjugated lipid nanoparticle/siRNA system inhibits androgen receptor expression in vivo[J]. Molecular Therapy Nucleic Acids, 2016, 5.
[4]. Zhou W, Huang J, Xiao Q, et al. Glu-Urea-Lys Scaffold Functionalized Superparamagnetic Iron Oxide Nanoparticles Targeting PSMA for In Vivo Molecular MRI of Prostate Cancer[J]. Pharmaceutics, 2022, 14(10): 2051.