Product Manual

Product Name	mRNA Cap 2′-O-Methyltransferase（GMP-grade）
Source	Recombinant expression in Escherichia coli
Catalog Number	CSB-DEM083
Physical Form	Liquid
Enzyme activity	50 U/μL
Storage Conditions	-20±5°C
Storage Buffer	20 mM Tris-HCl pH 8.0, 0.1 mM EDTA, 1 mM DTT, 100 mM NaCl, 50% (v/v) glycerol, 0.1% (v/v) Triton X-100.
Activity definition	The enzyme amount required to methylate 10 pmol of 80 nt capped RNA transcript within 1 hour at 37°C is defined as one unit (U).
Shelf Life	12 months

Product Description

mRNA Cap 2′-O-Methyltransferase is a recombinant protein derived from cowpox virus. This enzyme can add a methyl group at the 2′-O position of the first nucleotide adjacent to the cap structure at the 5′ end of RNA. The enzyme utilizes SAM as a methyl donor to methylate capped RNA, resulting in the formation of a Cap1 structure. Cap1 structure enhances mRNA translation efficiency, thus improving mRNA transfection and expression in microinjection experiments. The enzyme requires RNA with a m7GpppN cap structure as a substrate.

Product Components

Label	Components	Specifications		
1	mRNA Cap 2′ -O-Methyltransferase（GMP-grade）	10KU	50KU	250KU
2	10×Capping Reaction Buffer	0.4mL	2mL	10mL

Operating instructions

1. Dilute an appropriate amount of Capped RNA in RNase-free water to a final volume of 16 μL.
2. Heat the diluted RNA at 65°C for 5 minutes, then place it on ice for 5 minutes.
3. Set up the reaction system as shown in the table below (applicable for methylation reaction with Capped RNA up to 10 μg).

Component	Volume
Denatured Capped RNA	16 μL
10×Capping Reaction Buffer	2 μL
SAM (4 mM)	1 μL
mRNA Cap 2´-O-Methyltransferase (50 U/μL)	1 μL

4.Incubate at 37°C for 1 hour (for RNA fragments shorter than 200 nt, incubation time can be extended to 2 hours).

Precautions

1. The RNA used for the experimental reaction should be purified and dissolved in RNase-free water without any EDTA or salt ions.

2. It is recommended to heat at 65°C for 5 minutes before the reaction to remove secondary structures of the RNA. If the 5´ end structure of the transcript is complex, the heating time can be extended to 10 minutes.