Vaccinia Capping Enzyme GMP-grade V01

Product Description

Mature mRNA in eukaryotes carries a unique 5' cap structure (m^7GpppN), known as the methylguanosine cap, which is essential for translation initiation, protection from RNase degradation, and enhancement of mRNA stability during splicing and nuclear export. Vaccinia Capping Enzyme GMP-grade (VCE), developed in-house by Biori Biotech, integrates the three enzymatic activities required for capping: RNA triphosphatase, guanylyltransferase, and guanine-N7-methyltransferase. In the presence of S-adenosylmethionine (SAM) as a methyl donor and guanosine triphosphate (GTP), VCE directly and efficiently adds an m^7G cap to the 5' end of mRNA in the correct orientation, producing Cap0-capped RNA with up to 100% capping efficiency in a single reaction. Cap1 RNA can be obtained by using VCE in combination with 2'-O-Methyltransferase (2OM) (GMP-BP-E06).

Components

Component	GMP-BP-E05-1K	GMP-BP-E05-10K	GMP-BP-E05-100K
	1 KU	10 KU	100 KU
Vaccinia Capping Enzyme GMP-grade (10 U/μL)	0.1 mL	1 mL	10 mL
10× Capping Buffer	0.2 mL	2 mL	20 mL
GTP Solution (10 mM)	0.1 mL	1 mL	10 mL
SAM (32 mM)	20 μL	0.2 mL	2 mL

Storage

Store at -20±5℃.

Product Information

Product Name	Vaccinia Capping Enzyme GMP-grade
Source	Recombinant *E.coli*
Activity	10 U/μL
Unit Definition	One unit (U) is defined as the amount of enzyme required to incorporate 10 pmol of GTP into RNA in 1 hour at 37℃.
Capping Buffer	500 mM Tris-HCl (pH 8.0), 50 mM KCl, 10 mM $MgCl_2$, 10 mM DTT
Storage Buffer	20 mM Tris-HCl (pH 8.0), 100 mM NaCl, 1 mM DTT, 0.1 mM EDTA, 0.1% (v/v) Triton X-100, 50% (v/v) glycerol

Quality control

1. Solution appearance: clear and transparent, free of visible particulate matter.
2. Activity>10 U/μL.
3. Protein purity≥95%.
4. Free of exogenous DNase, RNase, exonuclease, and endonuclease activity.
5. Residual host-cell DNA: ≤100 pg/mg.
6. Residual host-cell protein: <50 ppm.
7. Heavy metals<10 ppm.
8. HBV, HCV, HIV, and mycoplasma: not detected.
9. Bacterial endotoxin: <5 EU/mL.
10. pH 7.5-8.5.

Recommended Capping System

Components	Volume
10× Capping Buffer	2 μL
GTP Solution (10 mM)	1 μL
SAM (2 mM)	1 μL
Vaccinia Capping Enzyme GMP-grade (10 U/μL)	1 μL
RNase Inhibitor GMP-grade (40 U/μL)	0.5 μL
Denatured RNA	50 pmol

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RNase-free ddH_2O	To 20 μL

*Incubate at 37℃ for 1 hour

Application

Cap 0 capping of mRNA for in vitro or in vivo translation. For Cap 1 structure, use in combination with mRNA Cap 2´-O-Methyltransferase GMP-grade (GMP-BP-E06).

Notes

1. Capping efficiency is influenced by the secondary structure at the RNA 5' end. It is recommended to denature the RNA prior to the reaction by heating at 65℃ for 5 min followed by immediate cooling on ice for 5 min.
2. The capping reaction is generally complete within 1 h. For RNA with complex 5'-end secondary structure or short RNA (≤200 nt), the incubation time may be extended to 2 hours.
3. SAM is a high-energy methyl donor with limited stability. Thaw SAM on ice and add it to the reaction mixture while keeping on ice throughout.
4. Reaction conditions, including enzyme amount and incubation time, should be optimized according to RNA length, secondary structure, and input quantity.
5. Vortex before use. Avoid freeze-thaw cycles.