PRODUCT INFORMATION

Vaccinia Capping Enzyme

v. 230602

Catalog number	C15037-500U
Package & Component	Name: Vaccinia Capping Enzyme (10,000 U/mL) — Amount: 500 U (1 vial) Name: 10X Capping Enzyme Reaction Buffer — Amount: 100 µL (1 vial)
Description	This capping enzyme contains three enzymatic activities (RNA triphosphatase, guanylyltransferase and guanine methyltransferase) to forming a Cap 0 structure (m7Gppp5'N) In the presence of the vaccinia capping enzyme, GTP and SAM (methyl donor) the In Vitro Transcripts mRNA can be efficient add capped in less one hour.
Source	*Escherichia coli*
Purity	>95% as determined by SDS-PAGE. Purified by Ni-NTA chromatography.
Unit Definition	One unit of Vaccinia Capping Enzyme is defined as the amount of enzyme required to incorporate 10 pmol of (α^{32}P) GTP into an 80 nt transcript in 1 hour at 37°C
Reaction Condition	1X Capping enzyme reaction buffer, supplemented with 0.5 mM GTP and 0.1 mM S-adenosylmethione (SAM). Incubate at 37°C. 10X Capping enzyme Reaction Buffer: 500 mM Tris-HCl (pH 8.0), 50 mM KCl, 10 mM $MgCl_2$, 10 mM DTT.
Storage Buffer	Capping enzyme is supplied in 20 mM Tris-HCl (pH 8.0), 100 mM NaCl, 1 mM DTT, 0.1 mM EDTA, 0.1% Triton® X-100 and 50% Glycerol.
Storage	Stored at -20°C. Avoid repeated freeze/thaw cycles.
Manuel	Capping procedures: 1. Combine RNA and nuclease-free H_2O to a final 15 µL. 2. Heating at 65°C for 5 minutes then chill on ice for 5 minutes. 3. Below reaction mixture should be prepared on ice and combined in the followir order:

Component	Amount	Final concentration
Denatured RNA	15 µL	-
10X Capping Buffer	2 µL	1X
10 mM GTP	1 µL	0.5 mM
SAM (2 mM)	1 µL	0.1 mM
Vaccinia Capping Enzyme	1 µL	10 U/rxn

	4. Gently mix the reaction thoroughly to achieve uniform distribution. 5. Incubate at 37°C for 30 minutes.
Notes	Reaction should be performed under RNase free condition. Use nuclease-free tubes, reagents and water to avoid RNase contamination. Also, wear gloves when working with RNA.

For Research Use Only. Not for use in diagnostic and therapeutic procedures.