



SMOBIO[®]

Small Bio, Smart Tool

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Product Information

EzRNA™ series

RNA Capping System

RC1000 50 RXN

| | |
|------------------------------------|--------|
| Vaccinia Capping Enzyme | 50 µl |
| 2'-O-Methyltransferase | 50 µl |
| 10X Capping Buffer | 100 µl |
| S-adenosylmethionine (SAM) (32 mM) | 50 µl |
| RNase Inhibitor (20 U/µl) | 50 µl |
| GTP (10 mM) | 50 µl |
| Nuclease-Free Water | 1 ml |

Storage

-20°C for 24 months

Description

The EzRNA™ RNA Capping System is a user-friendly product for post-transcriptional RNA modification. Both Vaccinia Capping Enzyme and 2'-O-Methyltransferase are included in the package, which are able to perform in a single reaction. The Vaccinia Capping Enzyme attach 7-methylguanylate cap (m7Gppp, Cap-0) to the 5' end of RNA to form m7Gppp5'-N-RNA (Cap-0 RNA). The 2'-O-methyltransferase utilizes Cap-0 RNA as a substrate, employing S-adenosine methionine (SAM) as a methyl donor to methylate 2' -OH of the first nucleotide at the 5' end of Cap-0 RNA, resulting in the formation of Cap-1 RNA.

Features

- 2'-O-Methyltransferase included for Cap-1 RNA
- High capping efficiency
- High stability
- RNase inhibitor included to enhance the stability of capping reaction.

Application

- Generation of 5' Cap-0 (m7Gppp) and Cap-1 (m7GpppNm-) RNA by enzymatic reaction
- mRNA synthesis for *in vitro* translation
- Gene expression studies
- mRNA vaccine development and therapeutics

Recommended protocol for preparing Cap-0 and Cap-1 RNA

■ Part 1 Heat-denature the RNA prior to capping reaction

| | |
|-------------------------|----------|
| Purified RNA (uncapped) | 10 µg |
| Nuclease-Free Water | to 13 µl |

Total volume 13 µl

Subject the sample to heat denaturation at 65°C for 5 minutes, then promptly proceed to incubate it on ice for another 5 minutes.

■ Part 2 Assemble the reaction for preparing cap-1 RNA

Add the following components in the order specified.

| | |
|---|-------|
| Denatured RNA (from part 1) | 13 µl |
| 10X Capping Buffer | 2 µl |
| GTP (10 mM) | 1 µl |
| SAM (4 mM, diluted from 32 mM stock) ^{*1} | 1 µl |
| Vaccinia Capping Enzyme | 1 µl |
| 2'-O-Methyltransferase | 1 µl |
| RNase Inhibitor | 1 µl |

Total volume 20 µl

Incubate at 37°C for 1 hour ^{*2,3}.

*¹Dilute adequate amount of 32 mM stock solution by Nuclease-Free Water prior to the reaction, and keep on ice until use.

*²For RNA less than 200 nt long increase incubation time to 2 hours.

*³If necessary, proceed with RNA purification for subsequent applications.

Other Information

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Caution: Not intended for human or animal diagnostic or therapeutic uses.

Related Products

| | |
|--------|---|
| CK1000 | Champion E. coli Transformation Kit |
| IT1000 | EzRNA™ T7 High Yield RNA Synthesis Kit, 50 RXN |
| IT1100 | EzRNA™ T7 High Yield RNA Synthesis Kit (Ψ -UTP), 50 RXN |
| IT1200 | EzRNA™ T7 High Yield RNA Synthesis Kit ($me^1\Psi$ -UTP), 50 RXN |
| TF1000 | SMO-HiFi DNA Polymerase, 100 U |
| TF3000 | G-HiFi DNA Polymerase, 100 U |
| TP1000 | ExcelTaq Taq DNA Polymerase, 500 U \times 1 |
| TP1200 | ExcelTaq 5X PCR Master Dye Mix, 200 RXN |
| TP5000 | ExcelTaq Hot Start II DNA Polymerase, 500 U |
| TQ1110 | ExcelTaq 2X Q-PCR Master Mix (SYBR, ROX), 200 RXN |
| TQ2110 | ExcelTaq 2X Q-PCR Master Mix (TaqMan, ROX), 200 RXN |
| DM2100 | ExcelBand 100 bp DNA Ladder, 500 μ l |
| NS1000 | FluoroVue Nucleic Acid Gel Stain (10,000X), 500 μ l |

The latest version of the manual can be downloaded from www.smobio.com/shop.

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