

## Description

Cas9 mRNA expresses a version of the *Streptococcus pyogenes* SF370 Cas9 protein (CRISPR Associated Protein 9). Cas9 functions as part of the CRISPR (Clustered Regularly Interspaced Short Palindromic Repeats) genome editing system. In the CRISPR system, an RNA guide sequence targets the site of interest and the Cas9 protein is employed to perform double-stranded DNA cleavage.

Cas9 mRNA encodes the Cas9 protein with an N and C terminal nuclear localization signal (NLS). The incorporation of two NLS signals within the mRNA increases the frequency of delivery to the nucleus, thus increasing the rate of DNA cleavage. Additionally, a C terminal HA epitope tag aids in detection, isolation, and purification of the Cas9 protein.

This mRNA is capped using CleanCap® Reagent M6, TriLink's patented co-transcriptional capping technology, resulting in the naturally occurring Cap-1 structure with >95% capping efficiency. It is polyadenylated, modified with N1-methylpseudouridine, and optimized for mammalian systems. It mimics a fully processed mature mRNA.

CleanCap® Reagent M6, otherwise known as CleanCap® m6AG 3'OMe, produces a base-modified Cap-1 mRNA, which shows superior *in vivo* activity compared to Cap-0 mRNA produced by legacy capping methods such as mCap

or anti-reverse cap analog (ARCA). CleanCap® Reagent M6 may further increase protein expression relative to previous generations of cap analogs, such as CleanCap® AG or CleanCap® AG (3'OMe), or mRNAs produced by enzymatic capping strategies<sup>1</sup>.

N1-methylpseudouridine is a modified uridine that can reduce immunogenic response and enhance translational efficiency of mRNAs. These properties can result in safer mRNA and increased protein expression.

Full length: 4,522 nucleotides  
ORF Length: 4,261 nucleotides

ORF sequence available online at [trilinkbiotech.com/cleancap-m6-cas9-mrna-n1mepsu.html](http://trilinkbiotech.com/cleancap-m6-cas9-mrna-n1mepsu.html)

CleanCap® M6 Cas9 mRNA (N1MePsU)<sup>S\*</sup> may be ordered using the following catalog numbers:

L-8106-100 (100 µg)  
L-8106-1000 (1 mg)  
L-8106-5 (5 x 1 mg)  
L-8106-BK (Bulk amount)

1.0 mg/mL in 1 mM sodium citrate, pH 6.4

Store at or below -40°C.

## Use & handling

Store at or below -40°C. Thaw and work on ice. Upon first use, pulse spin before opening and aliquot into single-use portions. Do not vortex. Use only certified RNase-free reagents and consumables with proper RNase-free technique. Use of barrier tips is recommended. Avoid freeze/thaw cycles. Do not mix with media containing serum unless first complexed with a stabilizing transfection reagent.

## QC analysis

- A260/A280 ratio
- dsRNA
- Concentration
- Fragment analyzer
- Agarose gel mobility
- Capping efficiency

A standard conversion factor of 40 µg/OD260 was used to calculate quantity.

Product released by Quality Assurance. TriLink is certified ISO 9001:2015.

## Troubleshooting

For any questions or technical support around this product, please reach out to [support@trilinkbiotech.com](mailto:support@trilinkbiotech.com)

<sup>1</sup>Final capping is dependent upon the CleanCap® Reagent, DNA template, and final mRNA sequence. Secondary structure due to RNA length and base composition can affect final capping efficiency, mRNA yield, and translation efficiency.

## Related TriLink and Alphazyme products

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CleanCap® M6 FLuc mRNA (N1MePsU) (cat no. L-8102)\*  
CleanCap® M6 mCherry mRNA (N1MePsU) (cat no. L-8103)\*  
CleanCap® M6 EGFP mRNA (N1MePsU) (cat no. L-8101)\*\*  
CleanCap® M6 EPO mRNA (N1MePsU) (cat no. L-8109)\*  
CleanCap® M6 Cre mRNA (N1MePsU) (cat no. L-8111)\*

CleanCap® OVA mRNA (cat no. L-7610)  
CleanCap® beta gal mRNA (cat no. L-7608)  
CleanCap® Cas9 mRNA (cat no. L-7606)<sup>§</sup>  
CleanCap® FLuc mRNA (cat no. L-7602)  
CleanCap® EGFP mRNA (cat no. L-7601)<sup>‡</sup>

CleanCap® Cre mRNA (5moU) (cat no. L-7211)  
CleanCap® OVA mRNA (5moU) (cat no. L-7210)  
CleanCap® EPO mRNA (5moU) (cat no. L-7209)  
CleanCap® beta gal mRNA (5moU) (cat no. L-7208)  
CleanCap® Cas9 Nickase mRNA (5moU) (cat no. L-7207)<sup>§</sup>  
CleanCap® Cas9 mRNA (5moU) (cat no. L-7206)<sup>§</sup>  
CleanCap® Renilla Luc mRNA (5moU) (cat no. L-7204)  
CleanCap® mCherry mRNA (5moU) (cat no. L-7203)

CleanCap® Fluc mRNA (5moU) (cat no. L-7202)  
CleanCap® EGFP mRNA (5moU) (cat no. L-7201)<sup>‡</sup>

CleanCap® Reagent M6 (cat. no. N-7453)  
CleanCap® Reagent AG (cat. no. N-7113)  
CleanCap® Reagent AG (3' OMe) (cat. no. N-7413)  
CleanCap® Reagent AU (cat. no. N-7114)

N1-Methylpseudouridine-5'-Triphosphate (cat. No. N-1081)\*  
5-Methoxyuridine-5'-Triphosphate (cat. no. N-1093)  
Pseudouridine-5'-Triphosphate (cat. No. N-1019)  
Nucleoside-5'-Triphosphate (NTP) Set (cat. no. N-1505)  
Adenosine-5'-Triphosphate, ATP (cat. no. N-1501)  
Cytidine-5'-Triphosphate, CTP (cat. no. N-1502)  
Guanosine-5'-Triphosphate, GTP (cat. no. N-1503)  
Uridine-5'-Triphosphate, UTP (cat. no. N-1504)

T7 RNA Polymerase (Alphazyme cat. No E057)  
Inorganic Pyrophosphatase (E. coli) (Alphazyme cat. No E051)  
Engineered RNase Inhibitor (Alphazyme cat. No E075)

## Related TriLink services

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TriLink offers RUO and GMP custom CleanCap® Cap-1 mRNA production services in addition to our catalog mRNA offerings. Visit our website [trilinkbiotech.com/mrna-cdm-services](http://trilinkbiotech.com/mrna-cdm-services) or contact us at [mrna-services@trilinkbiotech.com](mailto:mrna-services@trilinkbiotech.com) for more information.

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