Generate low immunogenicity N1meΨ-mRNA with transcript-to-tail production

INCOGNITO™ T7 mScript™ N1meΨ-mRNA Production System

Product Intro

The all-in-one, transcript-to-tail INCOGNITO™ T7 mScript™ N1meΨ-mRNA Production System synthesizes N1-methyl-pseudouridine-containing mRNA (N1meΨ-mRNA). It offers high-yield transcription incorporating N1meΨTP with post-transcriptional capping and tailing. The system achieves nearly 100% transcript capping efficiency with poly(A) tail lengths of >300 bases possible. Incorporating modified nucleosides into mRNA reduces innate immune responses in mammalian cells, making it ideal for transfection, microinjection experiments, and *in vitro* translation.

Benefits

- **All-in-one kit:** Eliminates the need for separate transcription, 5′ capping and 3′ tailing kits.
- **Lower immunogenicity:** Synthesized transcripts include N1meΨTP for reduced immune response.
- **High efficiency capping:** Achieve nearly ~100% 5′ capping.
- **Customizable poly(A) tails:** Generate variable 3′ poly(A) tail lengths, even >300 bases long.

Product Description

The INCOGNITO™ T7 mScript™ N1meΨ-mRNA Production System provides all enzymes and enzyme-related reagents for making N1meΨ-containing, 5′-capped, 3′-polyadenylated mRNA. It includes reagents for four workflow modules, (1) RNA transcription, (2) 5’ capping, (3) 3’ poly(A) tailing and (4) RNA/mRNA purification:

1. **Transcription:** *In vitro* transcription of linear double-stranded DNA templates using the T7 mScript™ Enzyme Solution with supplied N1meΨTP and canonical nucleotides ATP, CTP and GTP.
2. **5′ Capping:** Enzymatic capping of the RNA using ScriptCap™ Cap 1 Capping System (for making mRNA with a Cap 0 cap structure, or optionally making mRNA with a Cap 1 cap structure) (Figure 1).
3. **3′ Poly(A) Tailing:** A-Plus™ Poly(A) Polymerase for generating 3′poly(A) tails of 150-200 bases or, using a modified protocol, tail lengths >300 bases are possible.
4. **Purification:** 5M NH_4OAc is included as a convenient RNA and mRNA purification method.

Figure 1A: Cap 0-mRNA

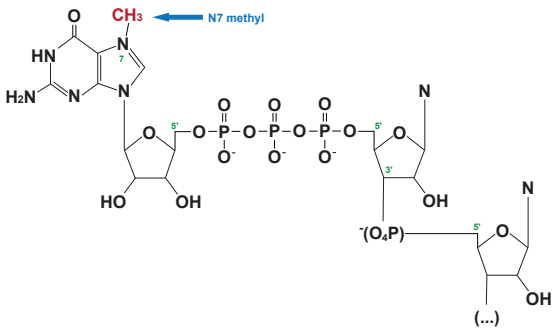

Figure 1B: Cap 1-mRNA

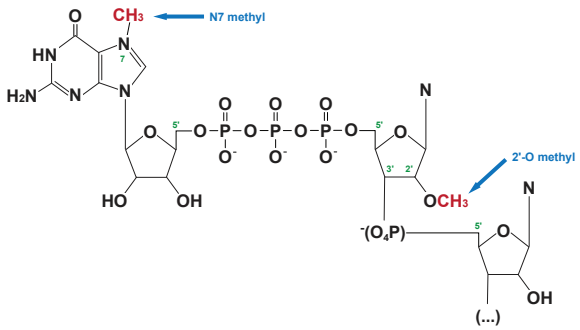

Figure 1: Cap 0-RNA and Cap 1-RNA. Cap 0 is added using ScriptCap™ Capping Enzyme and converted to Cap 1 using 2′-O-Methyltransferase.

For research use only

Post-transfection, capped and tailed mRNA has increased stability and translation efficiency in most eukaryotic cell lines. The mScript™ System improves upon existing capping methods by ensuring virtually 100% transcript capping, all caps in the proper orientation and the ability to produce large amounts of capped RNA at a reasonable cost. This mRNA is suitable for use in transfection and microinjection experiments as well as *in vitro* translation systems.

Performance Data

The INCOGNITO™ T7 mScript™ N1meΨ-mRNA Production System is functionally tested under standard reaction conditions using the T7 Control Template DNA. The *in vitro* transcription module must produce at least 50 µg of RNA from 1 µg of the T7 Control Template DNA in 20 minutes at 37°C. A-Plus™ Poly(A) Polymerase is functionally tested in 1X A-Plus™ Poly(A) Tailing Buffer with 1 mM ATP and a 1.4 kb transcript. The capping module enzymes are tested independently using non-T7 control transcript RNA to assay for completeness of reaction.

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Ordering information

Catalog Number	Description
IMMY240625	INCOGNITO™ T7 mScript™ N1meΨ-mRNA Production System (25 reactions)

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