

# Modified mRNA Products

#### Seporter Gene mRNA

APExBIO has developed and optimized mRNA reporters capped with ARCA or EZ Cap (equal to CleanCap) and polyadenylated by Poly A polymerase. These reporter mRNAs are transfected into mammalian cells as great controls to study the efficiency of transfection and translation through various assays (e.g. fluorescence microscopy, quantitative fluorometry, bioluminescent imaging & FACS).

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Cat.No.	Product Name	Information
R1007	ARCA EGFP mRNA (5-moUTP)	Direct-detection reporter mRNA that suppresses RNA-mediated innate immune activation, used as control to study transfection and expression in mammalian cells.
R1008	ARCA Cy3 EGFP mRNA (5-moUTP)	Cy3 tagged direct-detection reporter mRNA that suppresses RNA-mediated innate immune activation, used as control and tool for determining mRNA delivery and localization.
R1009	ARCA Cy5 EGFP mRNA (5-moUTP)	Cy5 tagged direct-detection reporter mRNA that suppresses RNA-mediated innate immune activation, used as control and tool for determining mRNA delivery and localization.
R1011	EZ Cap™ Cy5 EGFP mRNA (5-moUTP)	EGFP mRNA with Cap 1 structure (equal to CleanCap), modified by 5-moUTP and Cy5-utp, providing higher transcription efficiency and suppressing RNA-mediated innate immune activation.
R1012	ARCA Firefly Luciferase mRNA (5-moUTP)	Firefly Luciferase mRNA modified by ARCA and 5-moUTP, inhibiting RNA-mediated innate immune activation, stable and efficient expression efficiency, used as an experimental control.
R1010	EZ Cap™ Cy5 Firefly Luciferase mRNA (5-moUTP)	Firefly Luciferase mRNA with Cap 1 structure (equal to CleanCap), modified by 5-moUTP and Cy5-utp, providing higher transcription efficiency and suppressing RNA-mediated innate immune activation.
R1013	EZ Cap™ Firefly Luciferase mRNA (5-moUTP)	Firefly Luciferase mRNA with Cap 1 structure (equal to CleanCap), modified by 5-moUTP, providing higher transcription efficiency and suppressing RNA-mediated innate immune activation.

#### 🚱 Gene Editing mRNA

APExBIO supplies SpCas9 mRNA in an optimized version with ARCA or EZ Cap and poly-A tail. After transfected with guide RNA into cells, SPCas9 mRNA will be translated into the protein with high activity, recognizing and cleavage the targeted genomic DNA.

Cat.No.	Product Name	Information
R1006	SpCas9 mRNA (ARCA, 5me-CTP, ΨUTP)	Used with guide RNA for site-specific DNA cleavage in genome editing

#### **Reprogramming Gene mRNA**

mRNA-based reprogramming system has been thought as a reliable, safe and efficient tool of generating clinically relevant human iPS cell lines. Retrovirus used in traditional transduction of the reprogramming factors has the risk of viral integration in genome. APExBIO provides the several kinds of reprogramming factor mRNAs for safe and efficient iPS cell production.

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	Oct4 mRNA, human	KIf4 mRNA	Sox2 mRNA		c-Myc mRNA	



# Key Raw Materials for mRNA Synthesis

### Solution Modified Nucleotides

In vitro transcribed (IVT) mRNA is very sensitive to degradation by nucleases, which limits its suitability for transfections and therapeutic applications. The poly(A)-tail and the cap structure contribute to the stability of the mRNA. APExBIO supplies three types of capping products: mCAP, ARCA and EZ Cap (equal to CleanCap), to satisfy the varied needs. 5-Methyl-CTP, Pseudo-UTP and other modified nucleotides can also be incorporated into mRNA to reduce host cell immune response and enhances stability.

Cat.No.	Product Name	Cat.No.	Product Name	Cat.No.	Product Name
B8174	mCAP	B7967	5-Methyl-CTP	B8061	5-Methoxy-UTP
B8175	ARCA	B7972	Pseudo-UTP	B8178	EZ Cap <sup>TM</sup> Reagent AG $(3' \text{ OMe})$ (equal to CleanCap)

#### Second Second Labeled Nucleotides

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Cyanine dye-labeled or amine-labeled nucleotides can be incorporated efficiently into RNA transcripts via T7 polymerase for the direct visualization of the mRNA.

Cat.No.	Product Name	Cat.No.	Product Name
B7951	Aminoallyl-UTP	B8332	Fluorescein-12-UTP
B8330	Cy3-UTP	B8333	Cy5-UTP

#### Enzymes and Poly A Tail for mRNA Synthesis

mRNA can be synthesized by in vitro transcription (IVT) with the help of RNA polymerase. A poly (A) tail is added to the 3' end of the pre-mRNA once synthesis is complete. The poly (A) tail, combining with 5' cap protect the mRNA from degradation, assist in the export of the mature mRNA to the cytoplasm, and increase the translation efficiency. As IVT mRNA is very sensitive to degradation by nucleases, RNase Inhibitor is necessary for manipulating the mRNA in vitro.

Cat.N	lo. Product Nan	ne Cat.No.	Product Name	Cat.No.	Product Name
K1043	3 T7 RNA Polym	erase K1045	Poly A Polymerase	K1053	HyperScribe™ Poly (A) Tailing Kit
K1046	8 RNase Inhibito	K1047	HyperScribe™ T7 High Yield RNA Synthesis Kit		



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# Custom mRNA Synthesis

#### 🐅 In Vitro Transcription

Synthesis of mRNA transcripts containing modified nucleotides can be used for various biochemical and molecular biology studies. Large scale transcription reactions, generating up to 200 µg of mRNA per reaction are suitable for RNA amplification, expression studies (microinjection, infection with viral transcripts, in vitro translation), structural analysis (protein-RNA binding), and mechanistic studies (ribozyme analyses).



#### Solution Affordable Cost, High Yields

APExBIO offers affordable custom synthesis of mRNA and long RNA (up to multiple kilobases) with a wide array of modification services at scales ranging from micrograms to milligrams. The mRNA can be generated from DNA templates provided by our customers or we can provide a full service from the ground up. We provide mCAP, ARCA and EZ Cap (equal to CleanCap) capping or modified nucleotides implication for all our standard mRNA transcripts.

#### 🔈 Validation



EGFP mRNA in Hela cells; 24 hours post transfection



EGFP mRNA in PC3 cells; 48 hours post transfection



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3. Various modification, treatment and purification service

- Incorporates mCAP, ARCA and EZ Cap (equal to CleanCap) capping into the transcript to increase translation efficiency
- Reduces host cell immune response and enhances stability by incorporating modified nucleotides (5me-CTP, ψUTP and 5mo-UTP) and a poly-A tail
- Degrades the DNA template after mRNA synthesis with DNase
- Removes the 5' triphosphates at the end of mRNA with phosphatase to further reduce innate immune responses in mammalian cell
- Employs a robust clean-up spin column system and HPLC purification by ÄKTA Purifier that delivers high yields of mRNAs that are ready for most downstream applications

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#### Modified Nucleotides

Cat.No.	Product Name	Cat.No.	Product Name
B8174	mCAP	B7967	5-Methyl-CTP
B8175	ARCA	B7972	Pseudo-UTP
B8178	EZ Cap™ Reagent AG (3' OMe) (equal to CleanCap)	B8061	5-Methoxy-UTP



#### • theRNApeutics Platform

mRNA optimization for targeted cell or tissue requires large scale screening in vitro and in vivo. The high R&D cost hinders the most potential applications in disease treatment. *theRNApeutics* is a one-step, automatic mRNA platform from *RNACure*, taking advantage of core materials produced by APExBIO in screening and optimization.

#### 🐅 Features

- Start with target proteins in target diseases
- Design and optimize the sequence, the frame, and modification of mRNA
- Varied modified nucleotides for efficiency screening
- Screen and deliver mRNA into cultured cells, experimental animals for preclinical studies
- Build up clinical trials for promising mRNA drugs



Automated Workstation

We are partnering with *RNACure* for mRNA therapeutics

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