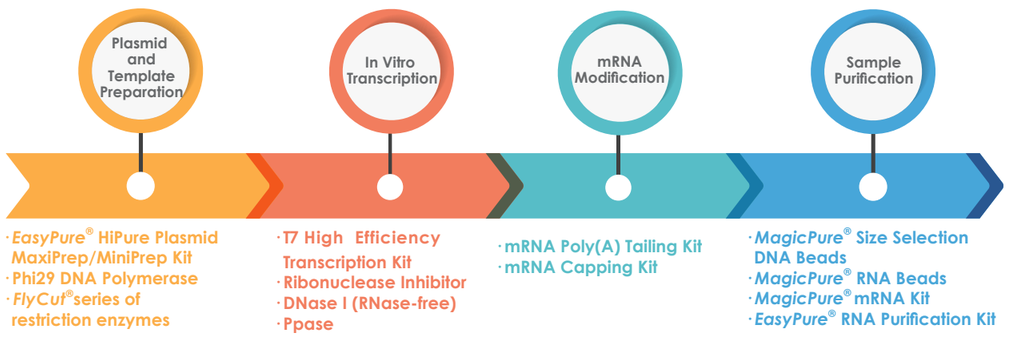
TRANSGEN BIOTECH

Reagents for In Vitro mRNA Synthesis

Providing Innovative Reagents for Life Sciences since 2006

mRNA plays an important role in biology, which can carry genetic information to guide the synthesis of proteins. With the launch and widespread application of COVID-19 mRNA vaccines, mRNA technology platforms have made major breakthroughs in 2020. mRNA has attracted much attention because of its advantages of safety and efficiency, and is a potential new therapeutic product. More and more companies are devoted to the research and development of mRNA drugs. The development process of mRNA drugs includes target gene selection, in vitro mRNA synthesis and modification, purification, LNP delivery and other operations. With 16 years of experience in molecular biology research and development, TransGen has developed a complete set of reagents for in vitro mRNA synthesis.

mRNA In Vitro Synthesis



Plasmid and Template Preparation

Plasmid DNA Purification

● *EasyPure*® HiPure Plasmid MiniPrep Kit (EM111)

•Visualized operationn: The color change indicates whether the lysis and neutralization are complete, thus ensuring the quality of plasmid extraction.
• Fast: Extraction completed within 1 hour.
• Simple: On-column endotoxin removal.
• High extraction volume.
• DNA is of high purity and free of endotoxins.

Fast restriction enzymes

● *FlyCut*®series of restriction enzymes

• Cut in 5 minutes.
• Universal buffer.
• No asterisk activity.

Isothermal Amplification

● Phi29 DNA Polymerase (LP101)

• Isothermal amplification.
• High Fidelity, High Efficiency, High Sensitivity, High Yield.
• Random or specific N9 primers can be used.

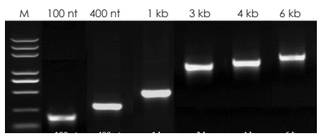
In Vitro Transcription

● T7 High Efficiency Transcription Kit (JT101)

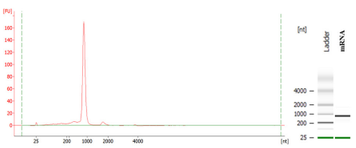
• High yield: >200 μg of RNA can be generated in a 20 μL reaction, and milligrams of mRNA can be prepared in a single 1 mL reaction.
• Good template compatibility: It is suitable for transcription of different types of DNA templates. The template input amount is 1 ng-2 μg, and the transcription fragment can reach more than 6 kb.
• High flexibility: The reaction volume can be scaled up or down according to experimental needs, compatible with cap analogs and chemically modified nucleotides.
• High quality: The product is of high purity and integrity.

Experimental Data

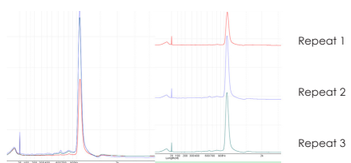
Good Transcription Specificity



The results of electrophoresis showed that the RNA products of different lengths were transcribed as a single band with good integrity;



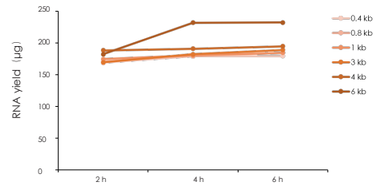
Agilent2100 detection results showed that the electrophoresis bands were uniform, the peak shape was single, and the product specificity was good.



The results of Qsep100 capillary electrophoresis showed that the product was single, without obvious non-target products, and of good integrity. The results of multiple experiments were reproducible.

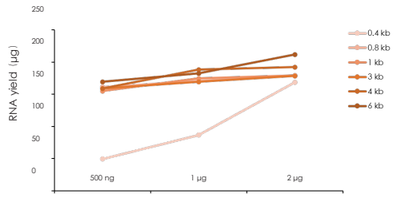
High Transcription Yield

RNA yield transcribed by 1 μg template



RNAs of different lengths were transcribed with 1 μg template, for transcription time of 2 h, 4 h, and 6 h, respectively. The results of concentration determination showed that a large amount of RNA could be obtained after 2 h of transcription.

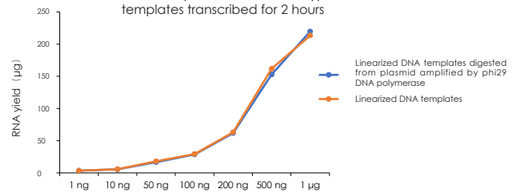
Transcription yield for 2 hours



RNAs of different lengths were transcribed with 500 ng, 1 μg, and 2 μg templates, respectively. Concentration assay results showed that TransGen products have high template compatibility.

Compatible with Different Types of DNA Templates

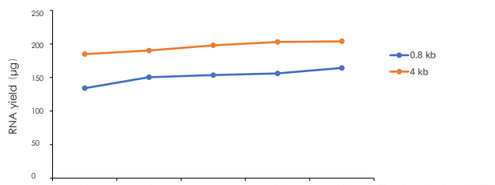
Yield comparison of different types of templates transcribed for 2 hours



The linearized DNA and linearized DNA digested from plasmid amplified by phi29 DNA polymerase were used as templates for in vitro transcription. The results showed that TransGen products were compatible with different types of DNA templates.

Compatible with Chemically Modified Nucleotides

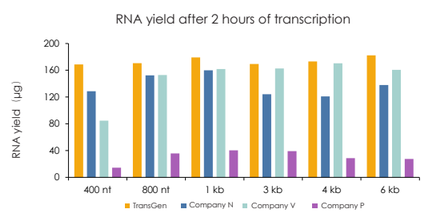
Transcription yield by 1 μg template for 2 hours



Replacing UTP with N1-Methyl-Pseudo-UTP in various ratios in the transcription reaction still yielded high RNA yields, indicating that TransGen products are compatible with chemically modified nucleotides.

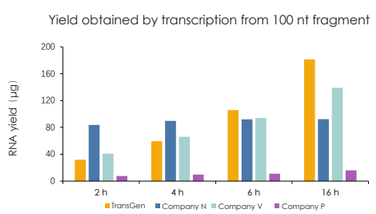
Comparison of Similar Products

RNA yield after 2 hours of transcription



Use TransGen and other similar products to transcribe DNA of different fragment lengths. The results showed that TransGen products have higher transcribed RNA yields than similar products.

Yield obtained by transcription from 100 nt fragment



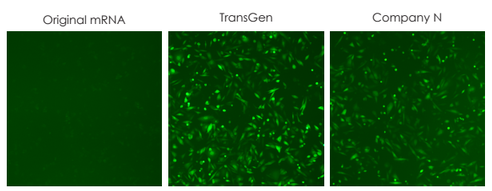
DNAs of 100 nt in length were transcribed using TransGen and other similar products respectively. The results showed that the RNA yield of TransGen products was higher than that of similar products after 6 h of transcription.

mRNA Modification

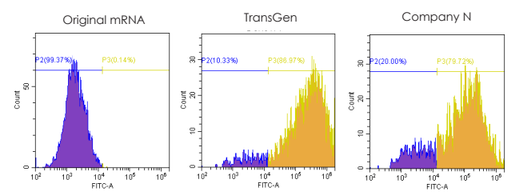
● mRNA Capping Kit (LC101)

• High efficiency: High capping efficiency and capping-modified mRNAs have high transfection and expression efficiency and good stability.
• Simple and quick: Add vaccinia virus capping enzyme and cap 2'-O-methyltransferase into the same reaction volume, and get Cap1 capped product within 1 hour.
• High flexibility: mRNAs of different lengths can be capped, and the reaction volume can be scaled up or down according to experimental needs.
• Provide a complete set of capping reaction raw materials: provide high-quality GTP and SAM as capping raw materials, and only need to prepare your own RNA for capping reaction.

Experimental Data



Fluorescence microscopy （48 HAT）

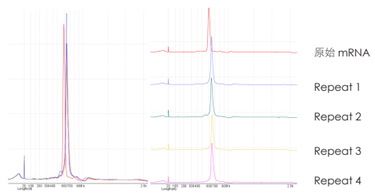


Transfection efficiency:0.14% Transfection efficiency:86.97% Transfection efficiency:79.72%
Flow cytometry （48 HAT）

The results of fluorescence microscopy and flow cytometry showed that the expression efficiency of capped mRNA using TransGen product (LC101) which was transfected into CHO cells was significantly higher than that of Company N product.

mRNA Poly(A) Tailing Kit (LA201)

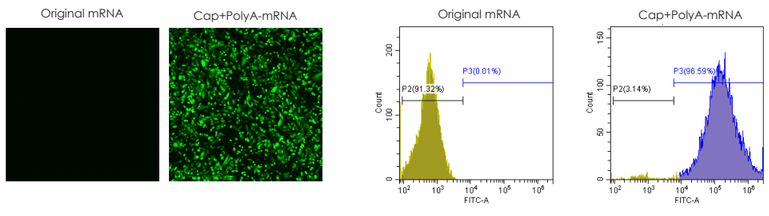
• High efficiency: The tailing efficiency is high and the length of Poly(A) tailing is relatively consistent.
• High flexibility: The reaction volume can be scaled up or down according to experimental needs.
• Quick and easy: The capped mRNA can be used directly in the TransGen product (LA201) for tailing reactions.
• Provide a complete set of tailing reaction raw materials: The reaction buffer contains the ATP raw materials required for tailing, and you only need to prepare your own RNA to carry out the tailing reaction.



The results of Qsep100 capillary electrophoresis showed that the peaks of RNA products using TransGen products were single and sharp, indicating uniform tail length and good stability.

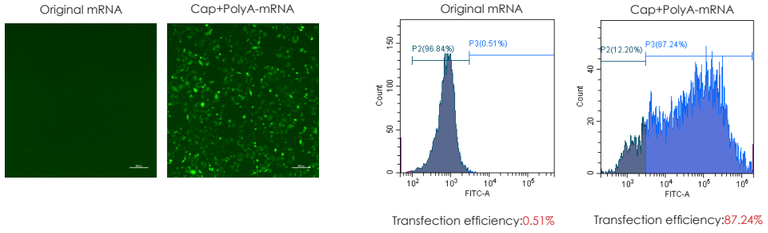
Complete mRNA Functional Data Presentation

CHO cells (Hamster)



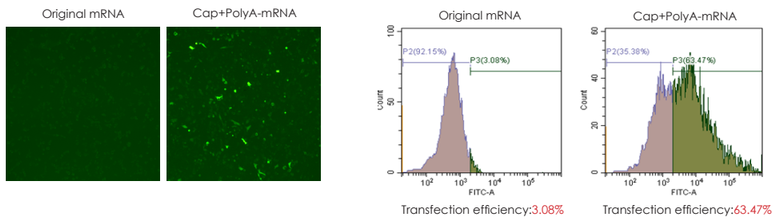
Transfection efficiency:0.01%　　Transfection efficiency:96.59%

Hela cells (Human)



Transfection efficiency:0.51%　　Transfection efficiency:87.24%

HEK293 cells (Human)



Transfection efficiency:3.08%　　Transfection efficiency:63.47%

The results of fluorescence microscopy and flow cytometry showed that the mRNA synthesized by TransGen mRNA in vitro synthesis products had low toxicity, good stability and high translation efficiency, and was successfully and stably expressed in different types of mammalian cells.

Sample Purification

DNA Purification

- ***MagicPure*® Size Selection DNA Beads (EC401)**

  Based on the Solid Phase Reverse Immobilization (SPRI) principle, a magnetic bead and buffer system with unique separation function is used, which is suitable for DNA purification, DNA concentration, and DNA fragment sorting in high-throughput sequencing library construction. The principle is compatible with mainstream library construction kits.

RNA Recovery Purification

- ***MagicPure*® mRNA Kit (EC511)**

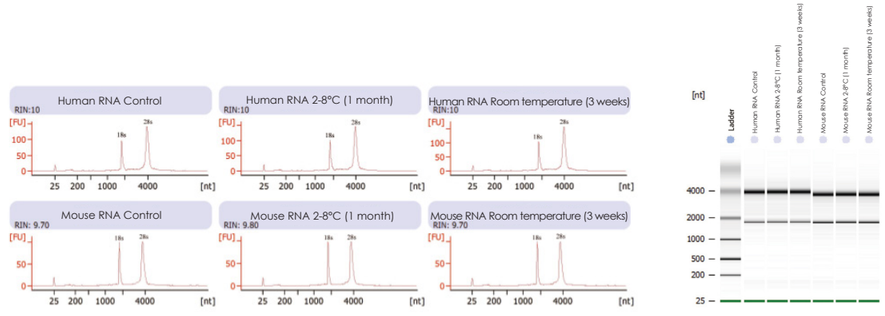
  Using Oligo (dT)-coupled magnetic beads to specifically bind to mRNA with Poly(A) tail, the purified mRNA has high purity and integrity, and this kit is suitable for magnetic rod type high-throughput nucleic acid extraction instrument.

- ***EasyPure*® RNA Purification Kit (ER701)**

  It can effectively remove impurities such as proteins, organic compounds, inorganic salt ions by using silica membrane spin column to specifically adsorb RNA and purify DNase I-treated total RNA products, in vitro transcription products, RNA-labeled products, synthetic RNA, etc. Operation is simple and fast.

- ***MagicPure*® RNA Beads (EC501)**

  Using a unique magnetic bead and buffer system, it can specifically adsorb RNA and purify RNA products from rRNA, DNase I-treated RNA products, in vitro transcription products, RNA-labeled products, and synthetic RNAs. Operation is simple and fast.



Control: Pure RNA samples　　2-8℃ (1 month): Purified RNA samples by EC501 after 1 month at 2-8℃
Room temperature (3 weeks): Purified RNA samples by EC501 after 3 weeks at room temperature

Agilent2100 test results show that TransGen products have good purification effect of human and mouse RNA and good product stability

Related Products

Category	Product Name	Cat. No	Specification
Plasmid and Template Preparation	*EasyPure*® HiPure Plasmid MiniPrep Kit	EM111-01	50 rxns
	EasyPure® HiPure Plasmid MaxiPrep Kit	EM121-01	10 rxns
	FlyCut® Fast Restriction Enzymes	See catalog	See catalog
	Phi29 DNA Polymerase	LP101-01	250 units
		LP101-02	5×250 units
In Vitro Transcription	T7 High Efficiency Transcription Kit	JT101-01	20 μl×25 rxns
		JT101-02	20 μl×100 rxns
	Ribonuclease Inhibitor	AI101-01	2000 units
		AI101-02	5×2000 units
	DNase I (RNase-free)	GD201-01	1500 units
mRNA Modification	mRNA Capping Kit	LC101-01	25 rxns
		LC101-02	100 rxns
	mRNA Poly(A) Tailing Kit	LA201-01	25 rxns
		LA201-02	100 rxns
DNA Purification	*MagicPure*® Size Selection DNA Beads	EC401-01	1 ml
		EC401-02	5 ml
		EC401-03	60 ml
		EC401-04	450 ml
RNA Purification	*MagicPure*® RNA Beads	EC501-01	1 ml
		EC501-02	5 ml
		EC501-03	60 ml
	MagicPure® mRNA Kit	EC511-01	24 rxns
		EC511-02	96 rxns
	EasyPure® RNA Purification Kit	ER701-01	25 rxns

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