

RNA Therapy Solutions

From Synthesis to Quality Control



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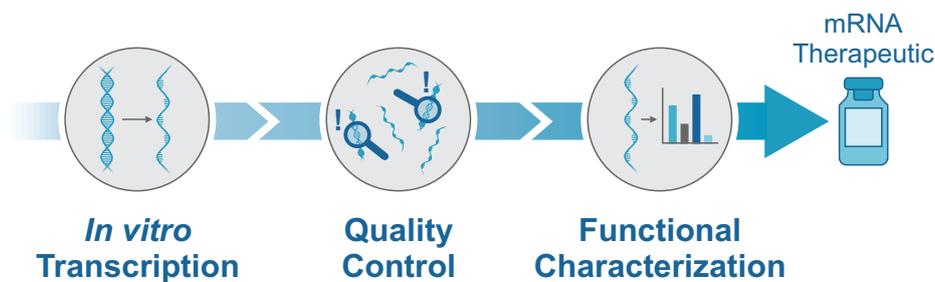
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1. Introduction

RNA therapeutics are an emerging area of research with increasing importance in modern medicine. They enable the development of treatments that act directly at the molecular level and can be tailored to specific biological mechanisms.

Promega offers a comprehensive portfolio supporting every stage of RNA therapeutic development, from plasmid preparation and *in vitro* transcription to quality control and functional characterization. The high-quality reagents ensure efficient and reliable RNA production. Analytical and functional assays such as dsRNA detection and TLR3 bioassays enable consistent and accurate quality assessment throughout the entire workflow.

With decades of experience in molecular biology and assay development, Promega provides reliable tools to support RNA research and therapeutic development.



From Transcription to Characterization – ensuring quality at every step.

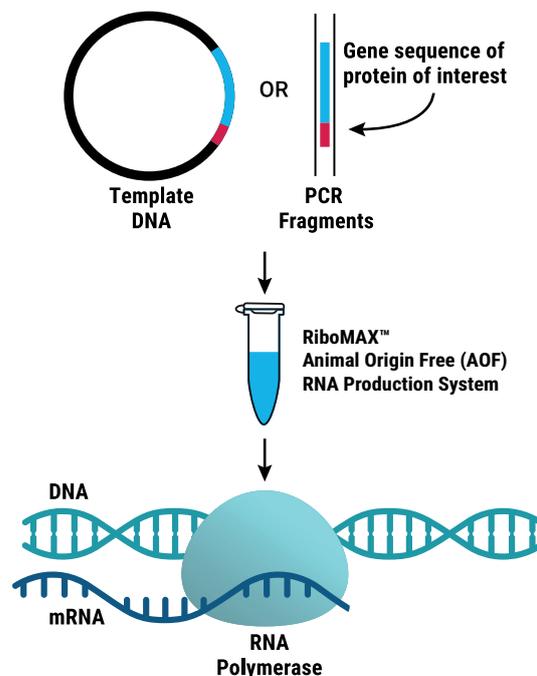
This diagram illustrates the main stages of the mRNA production workflow, including *in vitro* transcription, quality control, and functional characterization.

2. Comprehensive Solutions for mRNA Production

The choice of raw materials is critical for the success of mRNA manufacturing. High-quality and well-characterized components are essential for reproducibility, scalability, and regulatory compliance from early development to commercial production.

RiboMAX™ Animal Origin Free (AOF) RNA Production System

Designed for flexibility and performance, the kit provides all reagents required for efficient *in vitro* transcription. It includes individual components that can be adjusted to achieve optimal RNA yield and quality. Each reagent is animal-origin-free and cGMP grade. This design allows users to optimize conditions according to their template and process requirements. The reaction setup facilitates evaluation and refinement of parameters before transferring workflows to production scale. This results in consistent and reliable mRNA synthesis.



In vitro transcription of mRNA using the RiboMAX™ Animal Origin Free (AOF) RNA Production System

Key Features

- ✔ **Animal-origin-free, cGMP grade** – ensures safety, quality and compliance
- ✔ **Individually adjustable components** – enable tailored optimization
- ✔ **High-yield, fast workflow** – produces up to milligram quantities of mRNA in one hour
- ✔ **Refined enzyme mix** – preserves RNA integrity and consistent quality
- ✔ **Flexible use** – compatible with standard and modified nucleotides, including cap analogs
- ✔ **Scalable format** – smooth transition from small-scale to large-scale manufacturing
- ✔ **Convenient format** – provides reagents for five 1 ml reactions

What's Included in the Kit

(sufficient for 5 x 1 ml reactions or 50 x 100 µl reactions)

- ✔ T7 RNA Polymerase, AOF (400 U/µl)
- ✔ Inorganic Pyrophosphatase, AOF (2 U/µl)
- ✔ RNasin® Plus Ribonuclease Inhibitor, AOF (40 U/µl)
- ✔ 10 x Transcription Buffer (Magnesium-free)
- ✔ 1 M Magnesium Chloride (MgCl₂)
- ✔ 1 M Magnesium Acetate (Mg(OAc)₂)
- ✔ 1 M DTT
- ✔ rATP (100 mM)
- ✔ rGTP (100 mM)
- ✔ rCTP (100 mM)
- ✔ rUTP (100 mM)
- ✔ Nuclease-Free Water
- ✔ T7 Linear Control DNA

Ordering Information

Product	Size	Cat.#
RiboMAX™ AOF RNA Production System	5 x 1 ml	P2000

Tailored cGMP/AOF solutions are available in bulk, custom formulations, and flexible packaging options.

For more information or custom requests, please visit:
www.promega.com/customrawmaterials





Custom cGMP/AOF-Manufactured *In Vitro* Transcription Reagents

As you transition from research to production, the use of cGMP and animal-origin-free components becomes essential. Each reagent is supplied with data and comprehensive documentation to meet regulatory standards. All materials are manufactured under strict process controls to ensure consistent, high-quality results.

We offer a broad portfolio of compliant cGMP animal-origin-free raw materials to support mRNA manufacturing at any stage. The RiboMAX™ AOF System is designed for process development and utilizes the same high-quality raw materials that are also available as individual components. These components can be ordered separately and supplied in larger, custom fill formats for scale up and production. This ensures consistency, flexibility, and a reliable supply chain across all stages of mRNA manufacturing.

Research Use		cGMP for mRNA Therapeutics
✓	Function / Activity	✓
	Animal-Origin-Free	✓
	Contamination Testing For:	
✓	Nucleases	✓
	Bioburden	✓
	Host Cell DNA	✓
	Endotoxin	✓
	Documentation Available Upon Request:	
✓	Certificate of Analysis	✓
✓	Certificate of Origin	✓
	TSE / BSE Statement	✓

Key Features

- ☑ **Animal-origin-free** – manufactured without animal-derived components, using exclusively non-animal raw materials
- ☑ **Contamination testing** – minimizes the risk of impurities that could compromise RNA yield and quality
- ☑ **Comprehensive documentation** – CoA, CoO, and TSE / BSE statements available to support traceability and compliance

As a primary manufacturer, we control every step of production from raw materials to the finished reagent. Our cGMP/AOF *in vitro* transcription reagents are produced under a comprehensive quality system and manufactured according to ICH Q7 GMP principles. Integrated logistics support and global inventory locations ensure reliable availability and supply continuity. These reagents are offered in adaptable, scalable formats to support mRNA manufacturing from development to production.

Ordering Information

Product	Size	Cat.#
T7 RNA Polymerase	80 U/μl	P618X
	400 U/μl	P617X
RNasin® Plus Ribonuclease Inhibitor	40 U/μl	N373X
	500 U/μl	N372X
Inorganic Pyrophosphatase	2 U/μl	M934X
Proteinase K Solution	20 mg/ml	MC507X
Ribonucleotides		
rATP	100 mM	BX139X
rUTP	100 mM	BX140X
rGTP	100 mM	BX141X
rCTP	100 mM	BX142X
Enzyme Mix, T7 RNA Polymerase		
AOF version of Enzyme Mix contained in the T7 RiboMAX™ Large Scale RNA Production System (p/n P1300)		P337X
AOF version of Enzyme Mix contained in the T7 RiboMAX™ Express Large Scale RNA Production System (p/n P1320)		P199X

Tailored cGMP/AOF solutions are available in bulk, custom formulations, and flexible packaging options.

For more information or custom requests, please visit:
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3. Products for dsRNA Detection and Functional Analysis

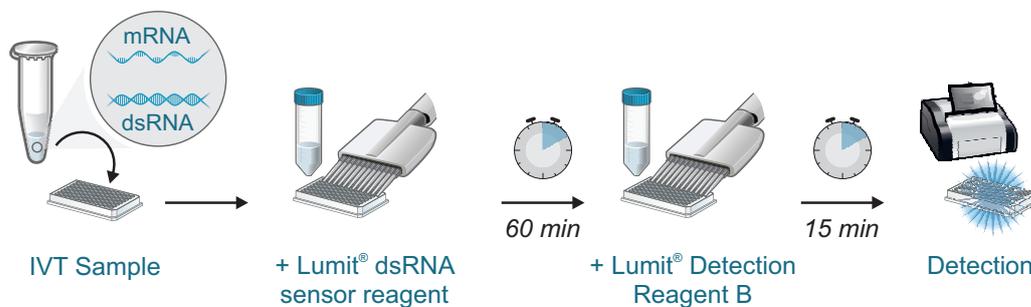
A common challenge in mRNA production is the formation of double-stranded RNA (dsRNA) during *in vitro* transcription (IVT). Even trace amounts of dsRNA can trigger unwanted immune responses, reducing the safety and effectiveness of mRNA therapeutics. Robust detection and control of dsRNA are therefore critical for maintaining consistent quality and meeting regulatory requirements in mRNA manufacturing.

Lumit[®] dsRNA Detection Assay

The Lumit[®] dsRNA Detection Assay provides a sensitive and straightforward solution for monitoring dsRNA contamination in mRNA preparations. Using NanoBiT[®] Luciferase complementation, a luminescent signal is generated only when dsRNA is present, enabling highly specific and quantitative detection. This simple, homogeneous workflow delivers consistent results within 75 minutes and supports robust process development and manufacturing.

Assay Workflow

The Lumit[®] dsRNA Detection Assay offers a fast and easy protocol with results in ~75 minutes. The workflow requires no wash steps and is fully compatible with standard microplate luminometers, making it ideal for high-throughput applications.



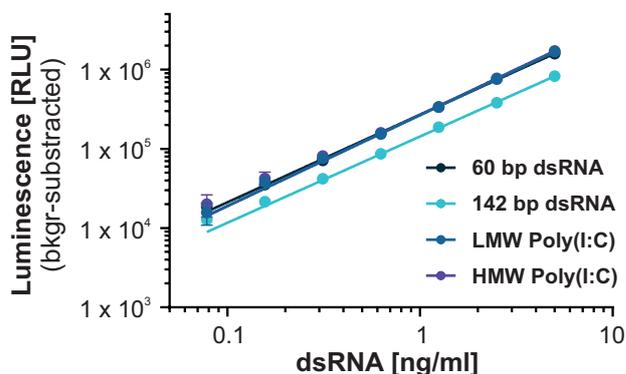
Fast and easy workflow: consistent results in just 75 minutes.

Key Features

- ☑ **High sensitivity** – detects dsRNA at low levels (0.04 – 2.5 ng/ml)
- ☑ **Rapid workflow** – results within 75 minutes, ideal for in-process testing
- ☑ **Simple and homogeneous** – no wash steps required, enabling easy implementation
- ☑ **Flexible format** – compatible with standard microplate luminometers for high-throughput analysis
- ☑ **Reliable QC tool** – supports mRNA manufacturing by ensuring product safety and consistency

Representative Data

Quantification across different dsRNA types



The Lumit[®] dsRNA Detection Assay sensitively quantifies dsRNA independent of sequence or size. Tested examples include short synthetic fragments (60 bp, 142 bp) and Poly(I:C) analogs of different lengths (LMW 0.2 – 1 kb; HMW 1.5 – 8 kb).

Ordering Information

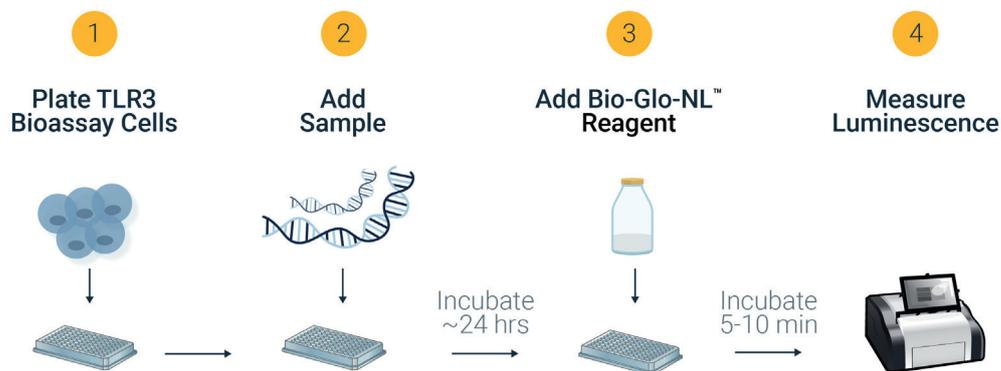
Product	Size	Cat.#
Lumit [®] dsRNA Detection Assay	100 assays	W2041
Lumit [®] dsRNA Detection Assay	500 assays	W2042
Standards with modified nucleotides:		
Lumit [®] dsRNA Standard, 5-Methylcytidine (100 µg/ml)	15 µl	CS355716
Lumit [®] dsRNA Standard, N1-Methylpseudouridine (100 µg/ml)	15 µl	CS355712
Lumit [®] dsRNA Standard, Pseudouridine (100 µg/ml)	15 µl	CS355710
Lumit [®] dsRNA Standard, 5-Methoxyuridine (100 µg/ml)	15 µl	CS355714

TLR3 Bioassay

The TLR3 Bioassay provides a functional readout of dsRNA-induced immune activation. It uses engineered cells expressing Toll-like receptor 3 (TLR3), which recognizes double-stranded RNA as a danger signal. Upon activation, TLR3 triggers a signaling cascade that induces a measurable reporter response. This assay enables direct evaluation of the biological activity of dsRNA contaminants, complementing quantitative detection methods such as the Lumit[®] dsRNA Detection Assay.

Assay Workflow

The TLR3 Bioassay uses engineered reporter cells that respond specifically to dsRNA stimulation. Following uptake into endosomes, dsRNA activates TLR3, triggering a signaling cascade and reporter expression measurable by luminescence. This simple, cell-based workflow generates biologically relevant results within ~24 hours.



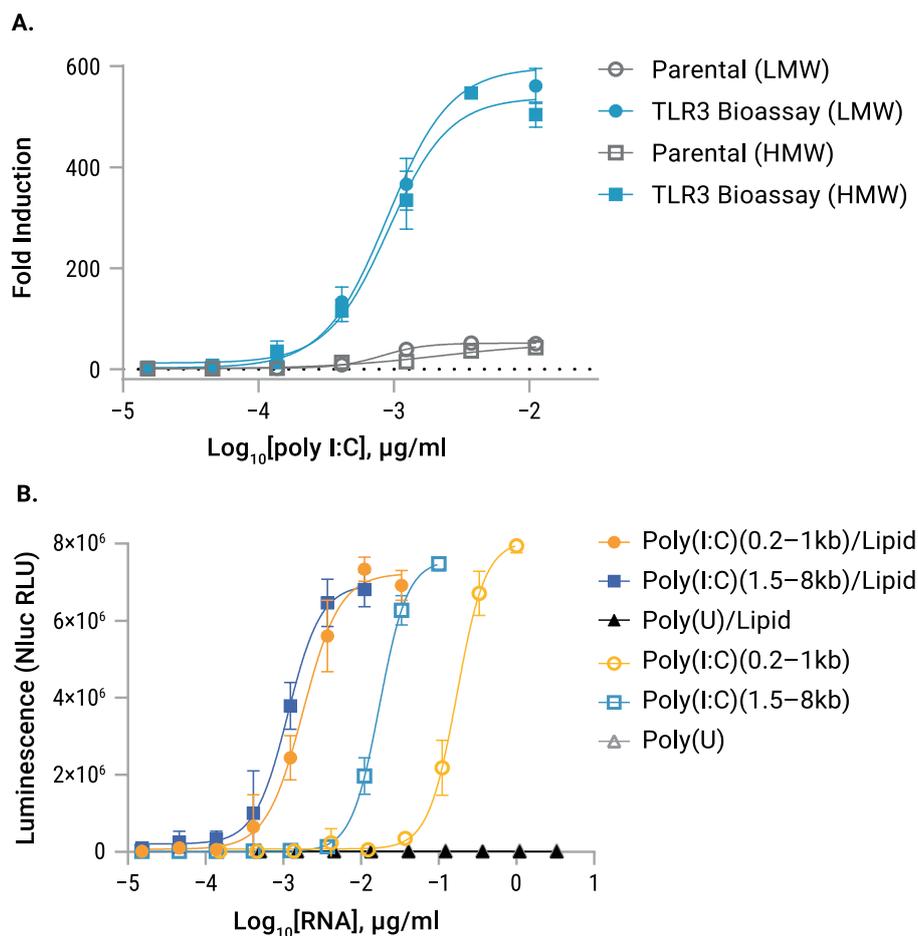
Cell-based TLR3 Bioassay delivers dsRNA detection within ~24 hours.

Key Features

- ✔ **Functional analysis** – measures dsRNA-induced TLR3 activation in a biologically relevant system
- ✔ **Sensitive and reproducible** – detects immune activation across a broad dsRNA concentration range
- ✔ **Ready-to-use format** – engineered cells and reagents provided, minimizing assay setup time
- ✔ **Quantifiable results** – reporter signal correlates with dsRNA activity for consistent assessment
- ✔ **Complementary tool** – pairs with Lumit[®] dsRNA Detection Assay for comprehensive mRNA quality control

Representative Data

Specific and sensitive detection of dsRNA-induced immune activation



Panel A. The TLR3 Bioassay specifically responds to dsRNA delivered with a transfection reagent, while parental reporter cells lacking TLR3 expression show no response. **Panel B.** The assay detects both low- and high-molecular-weight Poly(I:C), with or without transfection reagent, demonstrating broad applicability.

Ordering Information

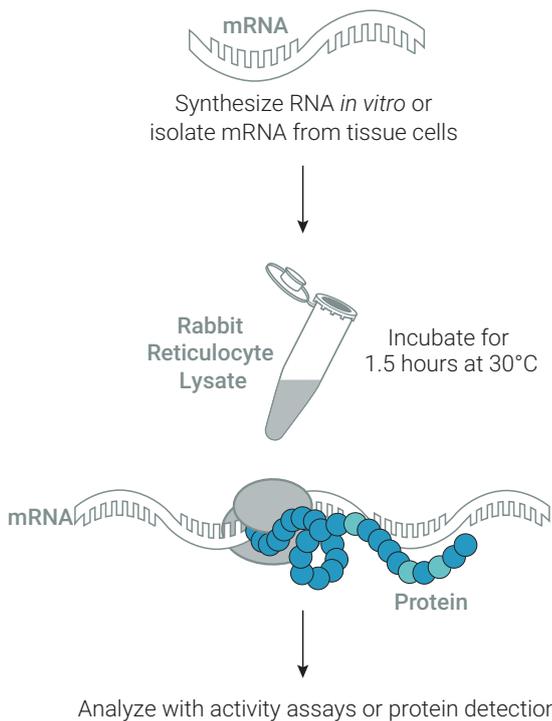
Product	Size	Cat. #
TLR3 Bioassay	1 x vial*	CS355505
	5 x vials*	CS355507

* Each vial of TLR3 Bioassay Cells is sufficient for 120 wells or two 96-well plates each in inner-60 well format.

4. *In vitro* Translation Lysates for mRNA Quality Assessment

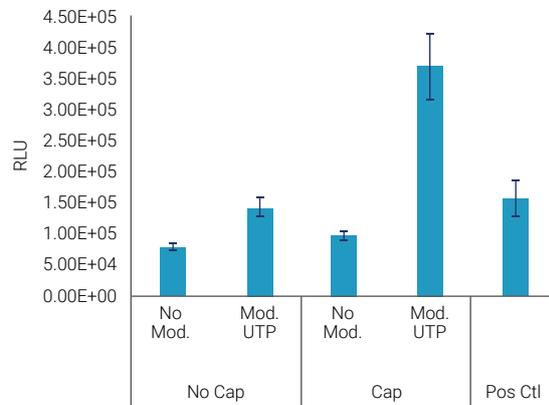
Cell-free lysates provide a fast way to evaluate mRNA quality and functionality. Rabbit Reticulocyte Lysate, Nuclease-Treated, and Wheat Germ Extract efficiently produce full-length proteins from mRNA templates. This includes capped and chemically modified transcripts. These systems deliver robust, reproducible tools for mRNA functional analysis.

For convenient, nonradioactive detection, Transcend™ tRNA incorporates biotin-labeled lysine during translation, enabling direct protein tagging. FluoroTect™ Green_{Lys} offers a fluorescence-based readout without labeling or antibodies, providing a simple and efficient option for real-time protein detection.



Protein expression using Rabbit Reticulocyte Lysate.

Representative Data



Effect of 5' capping and nucleotide modifications on *in vitro* protein production. Data show significantly increased protein production with capped RNA containing modified UTP compared to uncapped or unmodified controls.

Ordering Information

Product	Size	Cat.#
Rabbit Reticulocyte Lysate System, Nuclease Treated	30 reactions	L4960
Flexi® Rabbit Reticulocyte Lysate System	30 reactions	L4540
Wheat Germ Extract	30 × 50 µl reactions	L4380

5. Ordering Information

Products	Size	Cat.#
<i>In Vitro</i> Transcription		
RiboMAX™ AOF RNA Production System	5 x 1 ml or 50 x 100 µl	P2000
Animal-Origin-Free, cGMP-Manufactured, <i>In Vitro</i> Transcription Reagents		
T7 RNA Polymerase, 80U/µl	dispensed to order	P618X
T7 RNA Polymerase, 400U/µl	dispensed to order	P617X
T7 RNA Polymerase, 400U/µl (low Triton® X-100)	dispensed to order	BX260X
RNasin® Plus Ribonuclease Inhibitor, 40 U/µl	dispensed to order	N373X
RNasin® Plus Ribonuclease Inhibitor, 500 U/µl	dispensed to order	N372X
Inorganic Pyrophosphatase, 2 U/µl	dispensed to order	M934X
Proteinase K Solution	20 mg/ml	MC507X
Ribonucleotides (rATP)	100 mM, dispensed to order	BX139X
Ribonucleotides (rUTP)	100 mM, dispensed to order	BX140X
Ribonucleotides (rGTP)	100 mM, dispensed to order	BX141X
Ribonucleotides (rCTP)	100 mM, dispensed to order	BX142X
10 × Transcription Buffer (Magnesium-free)	dispensed to order	P211X
AOF version of Enzyme Mix contained in the T7 RiboMAX™ Large Scale RNA Production System (p/n P1300)		P337X
AOF version of Enzyme Mix contained in the T7 RiboMAX™ Express Large Scale RNA Production System (p/n P1320)		P199X
dsRNA Contamination		
Lumit® dsRNA Detection Assay	100 assays	W2041
Lumit® dsRNA Detection Assay	500 assays	W2042
Lumit® dsRNA Standard, 5-Methylcytidine (100 µg/ml)	15 µl	CS355716
Lumit® dsRNA Standard, N1-Methylpseudouridine (100 µg/ml)	15 µl	CS355712
Lumit® dsRNA Standard, Pseudouridine (100 µg/ml)	15 µl	CS355710
Lumit® dsRNA Standard, 5-Methoxyuridine (100 µg/ml)	15 µl	CS355714
TLR3 Bioassay	1 x vial	CS355505
TLR3 Bioassay	5 x vials	CS355507
<i>In Vitro</i> Translation		
Rabbit Reticulocyte Lysate, Nuclease-Treated	30 reactions	L4960
Flexi® Rabbit Reticulocyte Lysate, Nuclease-Treated	31 reactions	L4540
Wheat Germ Extract	30 × 50 µl reactions	L4380
Transcend™ Non-Radioactive Translation Detection System	30 reactions	L5070
FluoroTect™ Green _{Lys} in vitro Translation Labeling System	40 reactions	L5001
Transcend™ tRNA	30 µl	L5061

Legal Information

RiboMAX™ AOF RNA Production System, NanoBiT®, Lumit® dsRNA Detection Assay, Flexi® Rabbit Reticulocyte Lysate, Nuclease-Treated, Transcend™ Non-Radioactive Translation Detection System / tRNAFluoroTect™ Green Lysate, RNasin® Plus RNase and Bio-Glo-NL™ are trademarks or registered trademarks of Promega Corporation. Triton® X is a trademark of Union Carbide Corporation.

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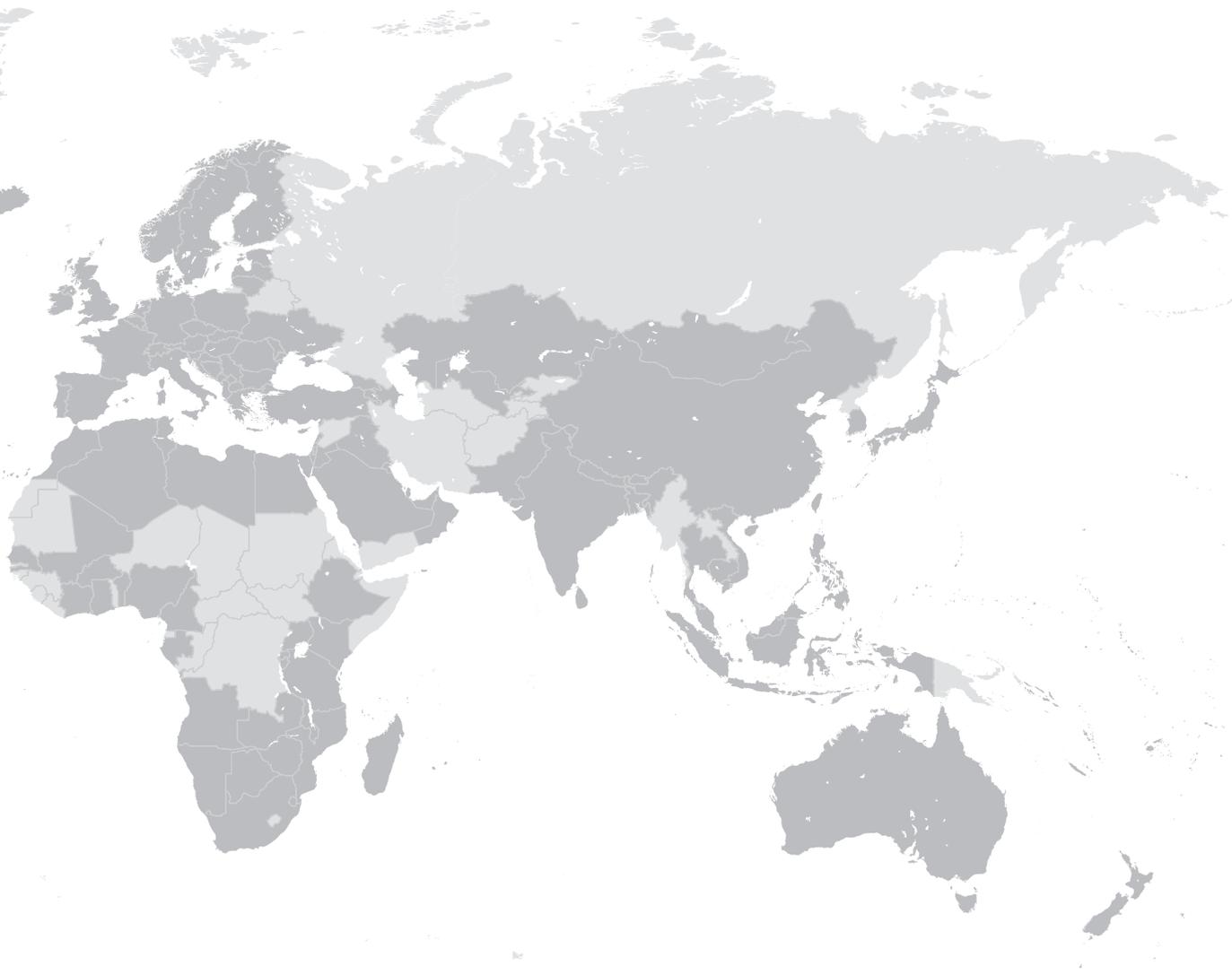
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