



AMPIGENE® Pre Lyo-Probe 1-Step Evaluation Kit

REF ENZ-NUC142-0200

200 Reaction

INTENDED USE:

For Research Use Only. Not for use in diagnostic procedures.

SUMMARY AND EXPLANATION

The **AMPIGENE® Pre-Lyo Probe 1-Step Evaluation Kit** is a version of the AMPIGENE® Lyo Probe 1-Step Kit that has been designed for evaluation purposes.

The kit includes a glycerol-free 4X qPCR mix containing hot start Taq polymerase, dNTPs, MgCl₂ and a blend of excipients to ensure reliable lyophilization, without loss of activity.

Lyo-Probe Reverse Transcriptase provided in this kit is for evaluation only. Due to its high glycerol content, it is not suitable for lyophilization. A highly concentrated version of Lyo-Probe Reverse Transcriptase is available as part of the **AMPIGENE® Lyo Probe 1-Step Kit** (ENZ-NUC143).

ASSAY PRINCIPLE

Polymerase chain reaction (PCR) uses *Taq* polymerase enzyme, which directs the synthesis of DNA from deoxynucleotide substrates on a single-stranded DNA template, by adding nucleotides to the 3' end of a custom-designed oligonucleotide annealed to the template DNA¹.

KNOWN APPLICATION

Amplification of nucleic acid targets with PCR methods.

PRODUCTS SUPPLIED

Component	200 reactions
AMPIGENE® Lyo-Probe Mix (4X)	1 x 1 mL
AMPIGENE® Lyo-Probe RTase (with RNase inhibitor) (20X)	1 x 200 µL

MATERIALS NEEDED (Not Provided)

- Target RNA
- Primers
- Thermal Cycler

STORAGE AND SHELF-LIFE

- Upon receipt, store kit at -20°C. These products are stable under these conditions up to the expiration date indicated in the vial label.
- Avoid prolonged exposure to light. If stored correctly the kit will retain full activity for 12 months from date of receipt. The kit can be stored at +4°C for 1 month. The kit can go through 30 freeze/thaw cycles with no loss of activity.

PERFORMANCE CONSIDERATIONS

1. Do not use reagents past their expiration date.
2. Cross-contamination of samples could cause false results. Use care when working with more than one sample.

LIMITATIONS

- This procedure is for research use only. It is not intended for diagnostic or therapeutic use.

PRECAUTIONS

1. Refer to reagent Safety Data Sheet (SDS) from precautions.
2. Specimens, before and after fixation, and all materials exposed to them should be handled and disposed of with proper precautions.
3. Never pipette reagents by mouth and avoid contact with skin and mucous membranes with reagents and specimens. If reagents and/or specimens come into contact with sensitive areas, rinse thoroughly with water and follow your institution's safety protocols.

TECHNICAL NOTES

For technical support and troubleshooting you can submit a technical enquiry online, call us direct, or alternatively email with the following information:

- Amplicon size
- Reaction setup
- Cycling conditions
- Screen grabs of gel images

GLOBAL HEADQUARTERS

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INSTRUCTIONS FOR USE

Template: The kit can be used with RNA extracted by most commercial kits, provided the amount and quality of template RNA are within an acceptable range.

Reaction Setup

1. Before starting, briefly vortex 4X AMPIGENE® Lyo-Probe Mix.
2. Prepare a master mix based on the following table. We also recommend setting up a no-RTase control:

Reagent	20 µL reactions	Final Concentration
AMPIGENE® Lyo-Probe Mix (4X)	5 µL	1x
Forward Primer (10 µM)	1-2 µL	400 nM – 1 µM
Reverse Primer (10 µM)	1-2 µL	400 nM – 1 µM
Probe (10µM)	0.25 – 1 µL	125 - 500 nM
AMPIGENE® Lyo-Probe RTase (20X)	1-2 µL	1x
RNA Template	2- 5 µL	Variable
PCR grade dH ₂ O	Up to 20 µL final volume	

3. Program the instrument using the following conditions, acquiring data on the appropriate channel:

Step	Temp. General	Temp. SARS-CoV-2 Detection	Time	Cycles
Reverse transcription	45°C to 55°C	55 °C	5-10 minutes singleplex	1
			10-20 minutes multiplex	
Polymerase activation and RTase inactivation	95°C	95 °C	3 minutes	1
Denaturation Anneal/Extension	95 °C	95 °C	15 seconds	50
	55 °C to 65 °C	58 °C	30 seconds	

REFERENCES

1. Mullis K, Faloona F, Scharf S, Saiki R, Horn G, Erlich H. Specific enzymatic amplification of DNA in vitro: the polymerase chain reaction. Cold Spring Harb Symp Quant Biol. 1986;51 Pt 1:263-73.

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