

Check the product label for actual catalog number, lot and expiry date.

SecurRIN™ Advanced RNase Inhibitor

CAT.#	SIZE	COMPONENTS	COMPONENT COMPOSITION
RNI0301	2500 u	1 x ~65 µl - SecurRIN™ Advanced RNase Inhibitor, 40 u/µl	Storage buffer contains 50% glycerol, 8mM DTT and other components.
RNI0305	12500 u	5 x ~65 µl - SecurRIN™ Advanced RNase Inhibitor, 40 u/µl	

Storage In the dark at -20°C.

APPLICATIONS

- Prevention of RNA degradation during:
 - cDNA synthesis
 - RNA extraction & storage
 - in vitro* transcription and translation
- Monoclonal antibody preparation procedures

DESCRIPTION

SecurRIN™ Advanced RNase Inhibitor is a premium tool for RNA protection from degradation during enzymatic reactions, storage or extraction. It is an extraordinary stable and robust enzyme: it works at up to 60°C temperature, remains active after weeks of room temperature exposure and multiple freezing thawing cycles. It is active in different buffer conditions within a broad pH range of 5.5 to 9.0, and within 0.5 - 1 mM concentration of DTT. SecurRIN™ Advanced RNase Inhibitor is a 50 kDa non-covalent inhibitor of RNase A, RNase B, and RNase C binding them in a 1 to 1 ratio. It is a recombinant protein derived from *E. coli* strain carrying human placenta RNase Inhibitor gene. SecurRIN™ Advanced RNase Inhibitor does not inhibit RNases H, 1, T1, T2 and S1 Nuclease. It influences neither the activity nor the performance of DNA polymerases and of Reverse Transcriptases. The enzyme is free from DNases and RNases.

BENEFITS

- Efficient RNase inhibition even at higher temperatures up to 60°C
- RNA protection from hydrolysis by RNases A, B and C
- Economical & stable for weeks at ambient temperature
- Robust, works well at 5.5 to 9.0 pH, with 0.5 - 1 mM DTT

PRODUCT DETAILS

- Active in all common buffers used for RNA work
- Acceptable pH range is 5.5 to 9.0
- Stable at ambient high temperature of up to 60°C
- Optimally performs at 0.5 - 1 mM DTT concentration
- 1 - 2 units of the RNase Inhibitor are typically enough for 1 microliter of RNA reaction. Enzyme amount depends on RNase contamination
- 1 µl of the enzyme is recommended for a standard cDNA synthesis reaction of 20 µl volume
- Binds and inhibits RNase A, RNase B and RNase C at 1:1 ratio
- Does not inhibit RNase H, RNase 1, RNase T1, RNase T2

UNIT DEFINITION

One unit is required to inhibit 5 ng of RNase A by 50% (measuring the hydrolysis of cytidine 2', 3'-cyclic monophosphate).

PROTOCOL example (please follow recommendations for Reverse Transcriptase you use)

- RNA is extremely sensitive to degradation by RNases present everywhere. Take care to protect RNA from degradation keep your bench clean, wear gloves, use sterile tubes and filter pipet tips.
- Check the integrity of RNA prior to cDNA synthesis in denaturing agarose gel.
- Add SecurRIN™ Advanced RNase Inhibitor in to the cDNA synthesis reaction (~40 units for 20 µl reaction tube) as the first component.
- In case high contamination with RNases is suspected, use 2 µl of the RNase Inhibitor. However, to avoid too much DTT in PCR, then use less of cDNA reaction mix as template for further PCR.
- Include positive and negative controls in parallel for each reaction.
- Thaw and keep reagents on ice. Mix well before use.
- For best results, optimize the template and primer amount.
- Choose optimal reaction temperature in a range recommended for your Reverse Transcriptase.

Perform reaction as recommended for your Reverse Transcriptase. Given temperatures and times are just approximate suggestions.

✓ Prepare a 20 µl reaction:	
SecurRIN™ Advanced RNase Inhibitor, 40 u/µl	1 µl (40 u)
10 mM dNTP Mix (NUM0201)	2 µl (1 mM final)
Total RNA <i>or</i>	1 pg to 5 µg <i>or</i>
Poly-A mRNA	1 pg to 0.5 µg
Oligo dT primer <i>or</i>	0.5 µg <i>or</i>
Random primer <i>or</i>	0.2 µg <i>or</i>
Specific primer	15-20 pmol
Water (PCR Water, WAT0110)	to 15 µl
✓ Mix gently, avoid bubbles.	
✓ Heat 5 min at 65°C, spin, place on ice for 1 min.	
✓ Add the 4 µl of 5X RT Reaction Buffer	
✓ Add 1 µl of Reverse Transcriptase, 200 u/µl and mix well.	
✓ Incubate 2 min at 42°C for Oligo dT and for Specific primer <i>or</i>	
10 min at 25°C for Random primer to anneal.	
✓ Incubate 30-50 min at 50°C to synthesize cDNA.	
✓ Inactivate at 85°C for 10 min.	
✓ Store reactions at -20°C or on ice for an immediate use.	
✓ Use 2-5 µl of this reaction mix per 50 µl PCR reaction.	
✓ Use 1-2 µl of this reaction mix per 20 µl qPCR reaction.	

IN VITRO RESEARCH USE ONLY

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