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RiboShield™ RNase Inhibitor

Product description

RiboShield™ RNase Inhibitor is a recombinant protein that blocks the activity of a wide range of ribonucleases to reliably protect your RNA from RNase digestion. The inhibitor is designed for use in RNA-sensitive applications such as RT-qPCR, cDNA synthesis and RNA-seq, where the presence of even small amounts of RNase can be highly detrimental to RNA quality and experimental outcome.

RiboShield™ RNase Inhibitor has a molecular weight of 50 kDa and is purified from High Five insect cells expressing a modified human placental gene. The inhibitor binds noncovalently to RNases at a 1:1 ratio, and has a Ki value of approximately 10^{-14} M when binding to RNase A[1]. Moreover, the very rapid kinetics of association to RNases guarantees immediate protection of your RNA.

Some cysteine residues present in human placental protein have been implicated in the oxidation sensitivity of the protein[2]. RiboShield™ RNase inhibitor does not contain these residues, resulting in a molecule more resistant to oxidative stress.

The high thermostability of RiboShield™ ensures activity up to 65 ˚C for 30 minutes. The inhibitor can block the activity of a wide range of ribonucleases, including eukaryotic RNases of the neutral type (e.g. RNases A, B and C). It does not inhibit RNases T1, T2, U1, U2, CL3, RNase I and H. The inhibitor is free from ribonucleases and phophatases, and is inactivated by heating at 75 ˚C for 15 minutes.

Component	2500 units	10000 units
RiboShield™ RNase Inhibitor (40 U/µL)	1 x 62.5 µL	4 x 62.5 µL

Shipping and storage

On arrival the kit should be stored between -30 ˚C and -15 ˚C. If stored correctly the kit will retain full activity for 12 months. Avoid multiple freeze-thaw cycles and exposing product to frequent changes in temperature

Limitations of product use

The product may be used for in vitro research purposes only.

Technical support

Help and support is available on our website at https://pcrbio.com/resources/ including answers to frequently asked technical questions. For technical support and troubleshooting you can submit a technical enquiry online, or alternatively email technical@pcrbio.com with the following information:

- Reaction type and setup,
- Cycling conditions,
- Screen grabs of gel images or amplification traces

Instructions for use

We recommend adding 40 units of RiboShield™ RNase inhibitor to a 20 µL reaction (1 µL per reaction to work with a final concentration of 2 U/µL). Titration may be required in case of templates derived from RNase-rich sources.

For RT-qPCR reactions, our kits already contain enough RiboShield RNase inhibitor to protect the RNA template in most of the cases. However, for templates derived from RNase-rich sources we recommend supplementing the reaction with additional 0.4 U/µL RNase inhibitor (i.e. adding extra 0.2 µL RiboShield RNase inhibitor to a 20 µL reaction).

RiboShield RNase inhibitor can be used to prevent RNA degradation after extraction (to prolong RNA viability during storage). In this case, we recommend using RiboShield RNase inhibitor as a 100x solution (i.e., the concentration of RNase inhibitor in the storage buffer should be 0.4 U/µL). Once again, higher amounts may be required in case of templates derived from RNase-rich sources.

RiboShield RNase inhibitor can also be used to block RNA degradation during RNA extraction in all those methods which do not include a protein denaturation step, given the proteic nature of the inhibitor itself. Also in these cases, the final concentration should be ≥0.4 U/µL.

References

[1] Lee FS, Shapiro R, Vallee BL. *Tight-binding inhibition of angiogenin and ribonuclease A by placental ribonucleasenhibitor.* Biochemistry. 1989; 28:225–230.

[2] Kim BM, Schultz LW, Raines RT. *Variants of ribonuclease inhibitor that resist oxidation.* Protein Science. 1999; 8(2):430-434.