

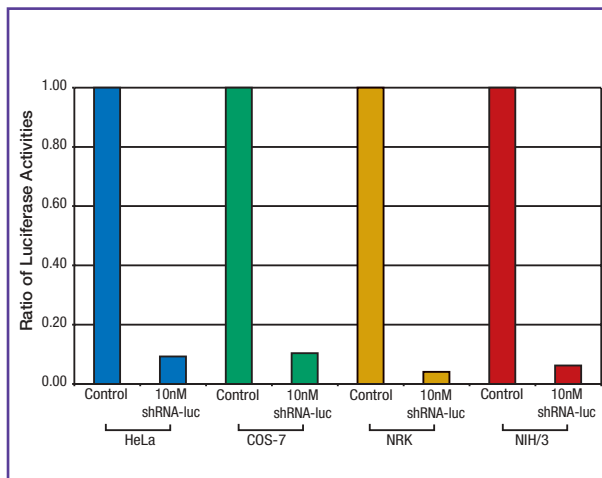
activity in the four cell types tested. At 10 nM, shRNA-luc specifically suppresses firefly luciferase expression by about 90% relative to *Renilla* luciferase expression in all cell types tested. Thus, the MessageMuter shRNAi Production Kit produces effective, target-specific shRNA.

The MessageMuter shRNAi Production Kit prepares short, dsRNA for RNAi studies

The MessageMuter™ shRNAi Production Kit uses *in vitro* transcription to prepare shRNA, which is an effective dsRNA for RNAi studies, in a rapid, simple, and cost-effective process. Each reaction produces enough shRNA, in 4 hours, for 100's of RNAi experiments.

Figure 5. shRNA-luc, produced with the MessageMuter™ shRNAi Production Kit, effectively induced sequence-specific silencing of firefly luciferase in a variety of cultured mammalian cells.

Transfections were performed in triplicate with firefly and *Renilla* (sea pansy) luciferase expression plasmids and 10 nM shRNA-luc, as described in the text. These experiments used the same shRNA-luc depicted in Figure 2C. Cells tested were HeLa (blue), COS-7 (green), Normal Rat Kidney (NRK; yellow) and mouse embryo fibroblast (NIH/3T3; red). Results for all experiments are shown as a ratio of firefly luciferase activity to *Renilla* luciferase activity.



References

- McManus, M.T. (2002) *RNA* **8**, 842.
- Paddison, P.J. et al. (2002) *Gene. Dev.* **16**, 948.
- Yu, J.-Y. et al. (2002) *Proc. Natl. Acad. Sci. USA* **99**(9), 6047.
- Meis, J.E. (2003) *EPICENTRE Forum* **10**(2), 1.
- EPICENTRE, unpublished observations.

www.epicentre.com/messagemuter.asp

MessageMuter™ shRNAi Production Kit

MM031110 10 Reactions

Each kit provides sufficient reagents to prepare 10 different shRNA. User supplies one DNA oligo per reaction.

Contents:
 MessageMuter™ T7 Promoter Oligo, 5X Annealing Buffer, Klenow exo-minus DNA Polymerase I, dNTPs, 10X Klenow Fill-In Buffer, AmpliScribe™ T7-Flash™ Enzyme Mix, AmpliScribe™ T7-Flash™ 10X Reaction Buffer, NTPs, DTT, Control Luciferase Oligo (60 bases), RNase-Free DNase I, Sterile Water.

Improve Your RT-PCR Results Using Thermostable MasterAmp™ Tth DNA Polymerase

In an RT-PCR reaction, secondary structure in the RNA template can cause premature termination and result in low yields of cDNA. MasterAmp™ Tth DNA Polymerase, derived from the thermophilic bacterium *Thermus thermophilus*, is a thermostable enzyme that has both DNA-directed DNA polymerase activity and efficient RNA-directed reverse transcriptase activity. The reverse transcriptase activity does not include RNase H activity. The enzyme has optimal activity at temperatures above 70°C and can be used at temperatures up to 95°C. Using Tth DNA Polymerase and high reaction temperatures minimizes problems due to nonspecific priming and RNA secondary structure. EPICENTRE's MasterAmp Tth DNA Polymerase is highly purified using a procedure that eliminates contaminating bacterial DNA.

MasterAmp Tth DNA Polymerase is supplied with the MasterAmp™ PCR Enhancer (with betaine)* which substantially increases both the ability to obtain

the desired amplification product and reaction-to-reaction consistency.

Applications of MasterAmp™ Tth DNA Polymerase

- High-temperature reverse transcription and RT-PCR.
- PCR amplification of DNA templates.

Benefits of MasterAmp™ Tth DNA Polymerase

- Optimal activity above 70°C eliminates mRNA secondary structure during reverse transcription and subsequent PCR reactions.
- Use the same reaction conditions for both thermostable reverse transcription and PCR. (1-tube, 1 enzyme RT-PCR)
- MasterAmp™ PCR Enhancer (with betaine) is supplied with the enzyme.

*Covered by issued and pending patents. This product is accompanied by a limited license to use in the Polymerase Chain Reaction (PCR) and RT-PCR for life sciences research in conjunction with a thermal cycler whose use in the automated performance of the PCR process is covered by the up-front license fee either by payment to Applied Biosystems or as purchased, i.e., an authorized thermocycler.

www.epicentre.com/tth.asp

MasterAmp™ Tth DNA Polymerase

TTH72100	5 U/μl	100 U
TTH72250	5 U/μl	250 U
TTH72500	5 U/μl	500 U
TTH7201K	5 U/μl	1,000 U
TTH7205K	5 U/μl	5,000 U (5 x 1,000 U)

Contents:
 20X PCR Buffer (without Mg²⁺ or Mn²⁺) plus separate 25 mM solutions of MgCl₂ and MnSO₄ and MasterAmp™ 10X PCR Enhancer.

MasterAmp™ Tth DNA Polymerase (Enzyme Only)

TTH7225N	5 U/μl	250 U
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For use with MasterAmp™ PCR PreMixes.

MasterAmp™ 10X PCR Enhancer

ME81201	1.5 ml
ME81205	5 ml
ME81210	10 ml

25 mM MgCl₂

FB4250	25 ml
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