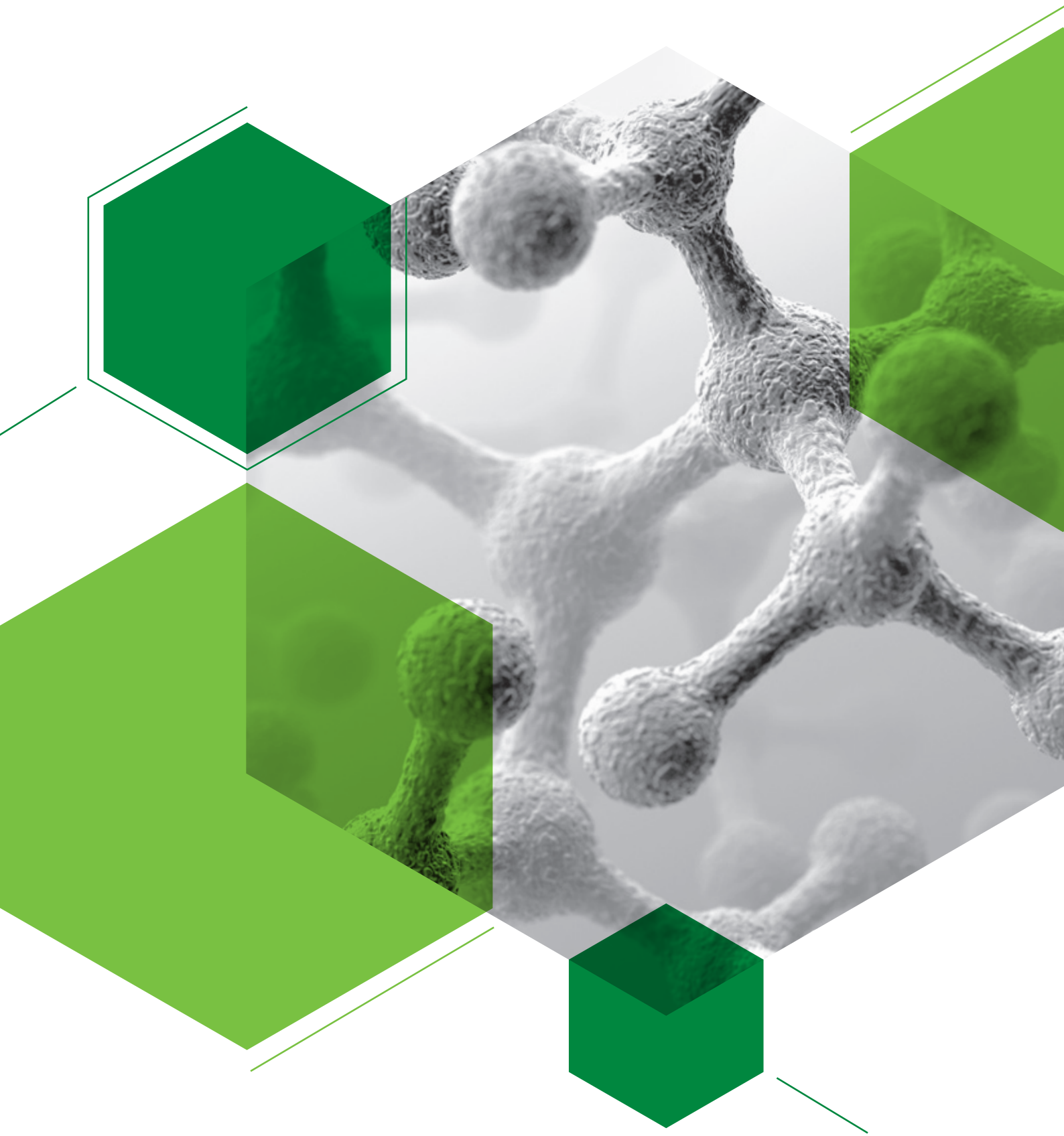
# RNA-related Products



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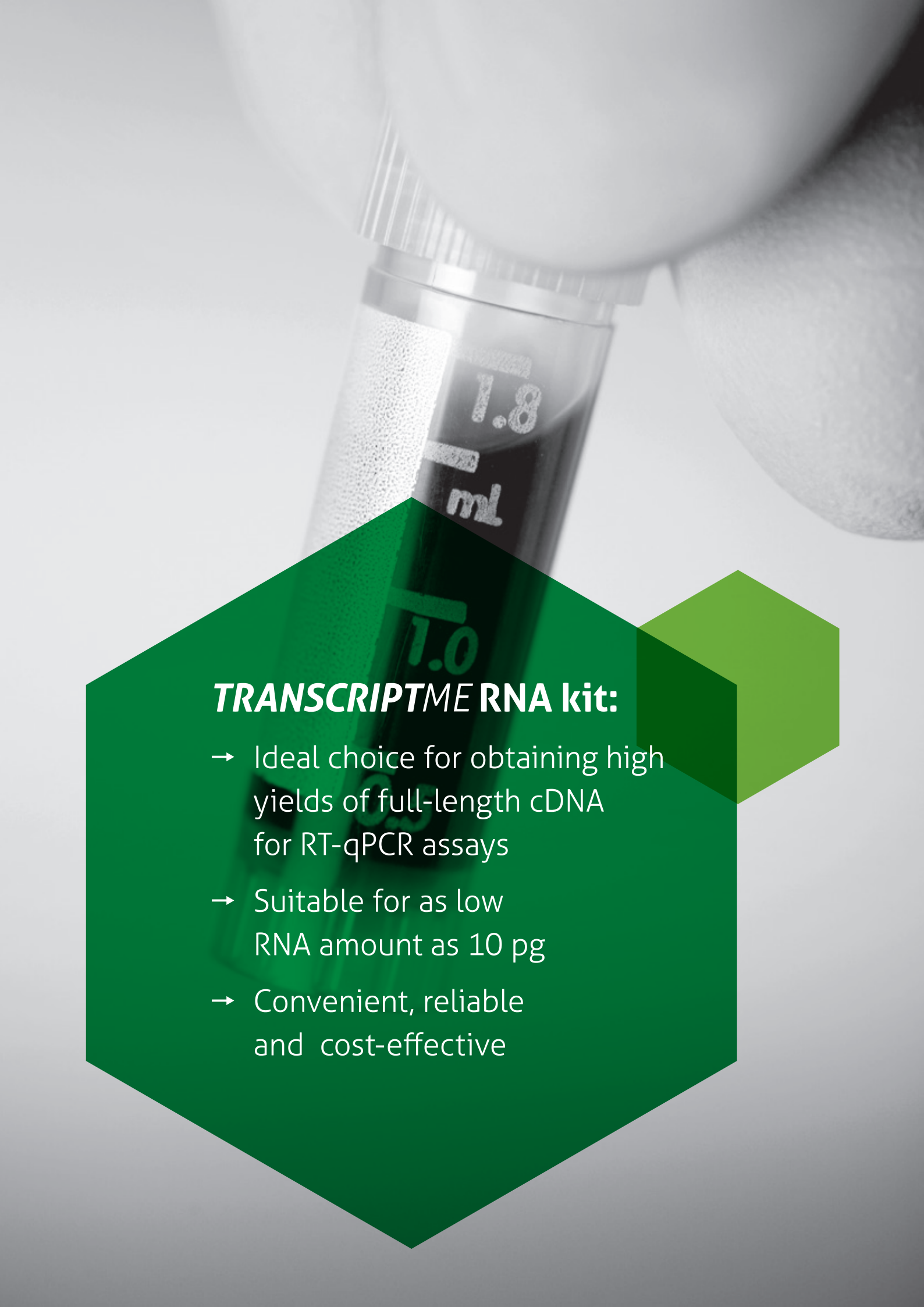



## ***TRANSCRIPTME*** **RNA kit:**

- → Ideal choice for obtaining high yields of full-length cDNA for RT-qPCR assays
- → Suitable for as low RNA amount as 10 pg
- → Convenient, reliable and cost-effective

# RNA-RELATED PRODUCTS

## *TRANSCRIPTME* RNA Kit – cDNA Synthesis Kit

The ***TRANSCIPTME*** **RNA Kit** is a system which includes all the necessary components to synthesize first-strand cDNA, except for the template RNA. The synthesized single-stranded cDNA is suitable for real-time quantitative RT-PCR applications. The ***TRANSCIPTME*** RNA Kit has been formulated to provide high yields of full-length cDNA product and to increase sensitivity in RT-qPCR. Starting material can range from 10 pg up to 5 µg of total RNA. The kit includes a combination of random hexamers and oligo(dT)$_{18}$ primers for increased sensitivity. The primers are included in the 2x Master Mix, which also contains dNTPs, $MgCl_2$ and an optimized RT buffer. ***TRANSCRIPTME*** Enzyme Mix includes both the ***TRANSCRIPTME*** **Reverse Transcriptase** (RNase H minus) and the ***RIBO****PROTECT* **RNase Inhibitor** for protection against RNA degradation caused by ribonuclease contamination.

The increased thermostability of the ***TRANSCRIPTME*** Reverse Transcriptase allows carrying out the reaction at a higher temperature (optimum activity at 50°C), which may increase the efficiency and specificity of the transcribed RNA regions, which are rich in GC pairs and/or contain secondary structures. The enzyme gives high yields of first strand cDNA up to 10 kb long.

**RNase H** (from *E. coli*) is provided as a separate tube in the kit to selectively degrade the RNA template in cDNA:RNA hybrids after the first-strand cDNA synthesis. This optional step can improve the sensitivity of subsequent RT-qPCR reaction since PCR primers will bind more easily to the cDNA. The protocol recommends using RNase H only when it contributes to a full-length cDNA synthesis and increased yields of first -strand cDNA.

**Features**

- High yields of full-length cDNA products (up to 10 kb)
- Formulated to increase sensitivity in RT-qPCR and RT-PCR assays
- Reduced number of pipetting steps – minimised contamination risk
- RNA starting material: 10 pg – 5 µg of total RNA or 10 pg – 500 ng of mRNA
- Optimal reaction temperature: 50°C
- Primer types: oligo(dT)$_{18}$ and random hexamers
- Suitable for the amplification of difficult RNA templates
- Convenient and reliable

**Applications**

- Full-length cDNA template synthesis for RT-qPCR and two-step RT-PCR assays
- cDNA synthesis for molecular cloning
- cDNA library construction
- RNA analysis

# The use of ***RIBO****PROTECT* **RNase Inhibitor** is highly recommended with samples containing endogenous RNases.

**Protect your RNA and avoid costly, unsatisfactory results.**

## ***TRANSCRIPT****ME* Reverse Transcriptase

The ***TRANSCIRPT****ME* is a modified, recombinant form of the Reverse Transcriptase from the Moloney Murine Leukemia Virus (M-MuLV) purified from *Escherichia coli*. The enzyme has been modified in order to promote stability. The ***TRANSCIRPT****ME* synthesizes the complementary DNA strand in the presence of a primer using either RNA (cDNA synthesis) or single-stranded DNA as a template. It lacks 3' → 5' exonuclease and RNase H activity, which improves synthesis of a full-length cDNA even from long mRNA templates, using random priming. The enzyme gives high yields of first strand cDNA up to 10 kb long.

The increased thermostability of the ***TRANSCRIPT****ME* allows carrying out the reaction at a higher temperature (optimum activity at 50°C), which may increase efficiency and specificity of the transcribed RNA regions which are rich in GC pairs and/or contain secondary structures.

**Concentration:** 200 U/µl

**Features**

- High yields of full-length cDNA synthesis (up to 10 kb long)
- Maintains the RNA- and DNA-dependent DNA polymerase activities
- Formulated to increase sensitivity in RT-qPCR and RT-PCR assays
- Starting material: 10 pg – 5 µg of total RNA or 10 pg – 500 ng of mRNA
- Optimal reaction temperature: 50°C
- Increased thermostability
- Lacks RNase H and 3'→ 5' exonuclease activities
- Suitable for the amplification of difficult RNA templates

**Applications**

- Full-length cDNA synthesis for use in RT-qPCR and two-step RT-PCR assays
- cDNA synthesis for molecular cloning
- cDNA library construction
- RNA analysis

## ***RIBO****PROTECT* – RNase Inhibitor

The ***RIBO****PROTECT* RNase Inhibitor is a recombinant inhibitor of pancreatic ribonucleases, such as RNase A, RNase B and RNase C, purified from *Escherichia coli*. This protein is useful in any application where eukaryotic RNase contamination is a potential problem. This inhibitor can be used to protect RNA template in cDNA synthesis or *in vitro* transcription/translation reactions. The ***RIBO****PROTECT* is not effective against RNase 1, RNase T1, RNase T2, S1 nuclease, RNase H or RNase from *Aspergillus sp.*

**Applications**

- cDNA synthesis, RT-PCR and RT-qPCR
- *in vitro* transcription/translation
- RNA extraction and purification

**Concentration:** 40 U/µl

# *EXTRACTME* TOTAL RNA KIT

The ***EXTRACTME*** **TOTAL RNA** kit is designed for the rapid and efficient purification of high quality RNA from 1-30 mg of tissue (fresh or frozen) and $10^4$-$10^7$ cultured cells.

**Product specifications:**

## SAMPLE MATERIAL

- fresh or frozen tissue (stored at -80°C): 1-30 mg
- tissue preserved in RNase inactivating buffers: 1-30 mg
- cell culture: $10^4$-$10^7$ cells
- body fluids (urine, cerebrospinal fluid, peritoneal fluid): 1-5 ml
- hair: 1-30 mg

## BINDING CAPACITY

- Approx. 90 µg RNA

## TIME REQUIRED

- 16-20 minutes (lysis and homogenisation time not included)
- 30-60 minutes for homogenisation in liquid nitrogen
- 30-40 minutes for mechanical homogenization (ceramic beads)

## RNA PURITY

- $A_{260}/A_{280}$ ratio = 1.9 – 2.1

**Average RNA isolation efficiencies from fresh biological material:**

| Sample material | Mass /quantity | Elution volume | RNA conc. | $A_{260}/A_{280}$ | Yield |
|---|---|---|---|---|---|
| **HCT116 cell culture** | $10^7$ | 100 µl | 947.2 ng/µl | 2.09 | 94.72 µg |
| **HCT116 cell culture** | $10^4$ | 100 µl | 328.3 ng/µl | 2.03 | 32.83 µg |
| **Liver** | 30 mg | 100 µl | 923.6 ng/µl | 2.07 | 92.36 µg |
| **Liver** | 10 mg | 100 µl | 319.3 ng/µl | 1.88 | 31.93 µg |
| **Liver tumor** | 30 mg | 100 µl | 534.6 ng/µl | 2.04 | 53.46 µg |
| **Liver tumor** | 15 mg | 100 µl | 467.9 ng/µl | 1.91 | 46.79 µg |
| **Colon** | 10 mg | 100 µl | 168.8 ng/µl | 2.14 | 16.88 µg |
| **Colon tumor** | 30 mg | 100 µl | 603.7ng/µl | 2.06 | 60.37 µg |

## RNase H

The RNase H is a recombinant endoribonuclease purified from an *E. coli* strain that carries the cloned RNase H gene (*rnh*). The enzyme selectively hydrolyses the phosphodiester bonds of RNA only when it is hybridized to DNA. The RNase H does not degrade single and double-stranded DNA or unhybridized RNA. It is a key enzyme in the removal of mRNA after first-strand cDNA synthesis. In addition, RNase H is useful for the removal of poly(A) tails on mRNAs after hybridization with oligo(dT), and for oligodeoxyribonucleotide-directed site-specific cleavage of RNA.

**Concentration:** 5 U/µl

## RNase A (DNase-free)

The RNase A is an endoribonuclease, that selectively cleaves single-stranded RNA 3' next to pyrimidine residues (cytosine, uracil). It degrades RNA to cyclic nucleotide monophosphates leaving a 5'-OH and 2'-, 3'-cyclic monophosphate. The enzyme exhibits no endonuclease or exonuclease activity towards DNA substrates. The RNase A is used to remove RNA during the isolation procedures of plasmid and genomic DNA.

**Applications**

- RNA protection assays
- Purification of RNA-free DNA
- Plasmid and genomic DNA isolation
- Removal of RNA during recombinant proteins preparations

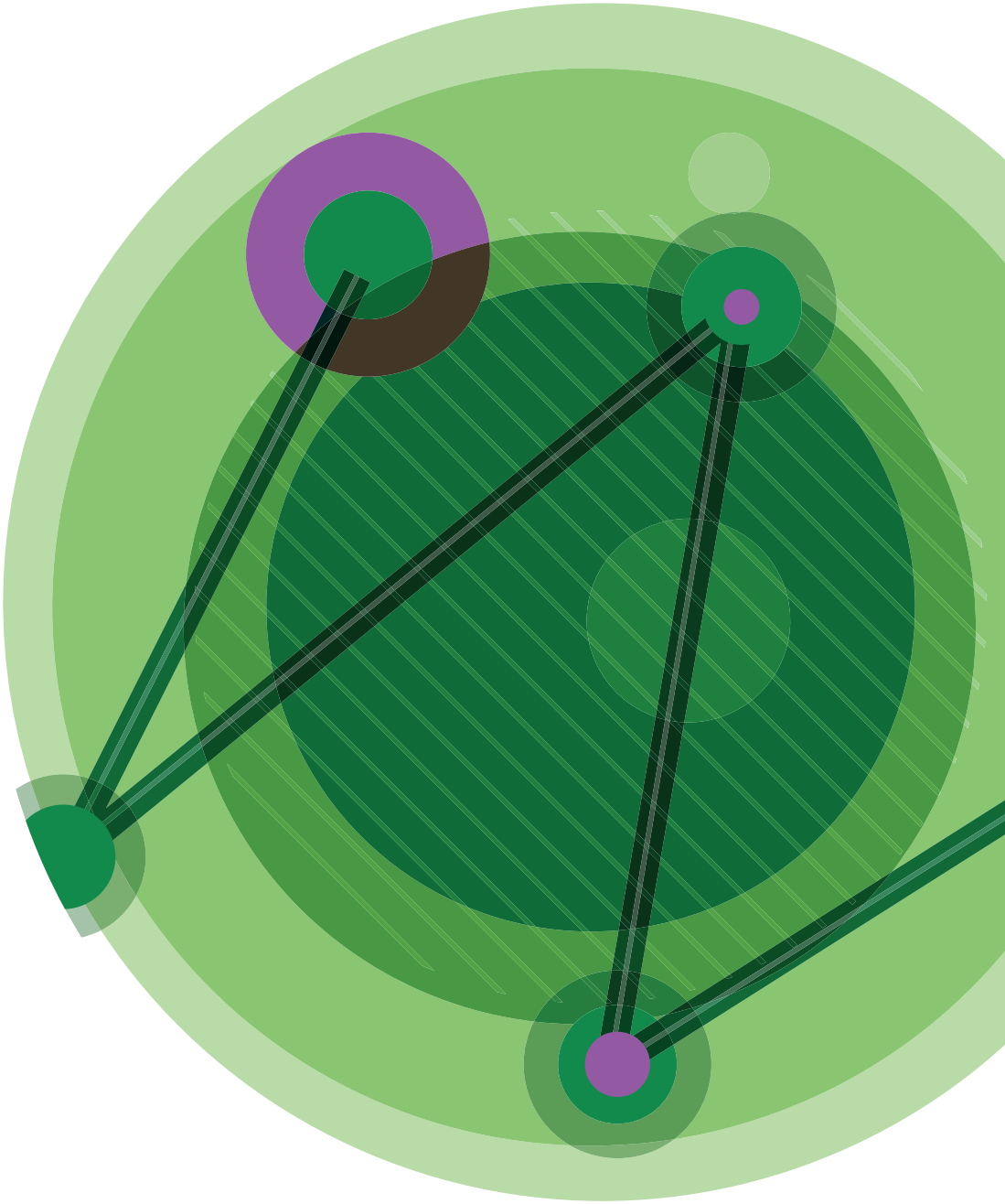
**Activity:** 90 U/mg (Kunitz)

| PRODUCT | SIZE | CAT. NO. |
|---|---|---|
| ***TRANSCRIPT****ME* **RNA Kit** cDNA synthesis Kit | 20 reactions | RT31-020 |
| | 100 reactions | RT31-100 |
| ***TRANSCRIPT****ME* Reverse Transcriptase | 10.000 U | RT32-010 |
| | 50.000 U | RT32-050 |
| ***RIBO****PROTECT* RNase Inhibitor | 2000 U | RT33-020 |
| | 10.000 U | RT33-100 |
| ***EXTRACT****ME* TOTAL RNA KIT | 25 isolations | EM09-025 |
| | 100 isolations | EM09-100 |
| | 250 isolation | EM09-250 |
| ***EXTRACT****ME* TOTAL RNA PLUS KIT The kit additionally include tubes for homogenisation with ceramic filling | 25 isolations | EM11-025 |
| | 100 isolations | EM11-100 |
| | 250 isolation | EM11-250 |
| **RNase H** | 250 U | RT34-025 |
| | 1250 U | RT34-125 |
| **RNase A** (DNase-free) | 50 mg | RP145 |

# EXTRACTME®
## TOTAL RNA KIT

The first step to successful RNA analysis is the extraction of pure, intact and high-quality RNA.

We recommend the ***EXTRACTME*** **TOTAL RNA KIT** for extraction of high quality RNA from animal tissue or cell cultures.

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