

GeneRead™ Pure mRNA Kit

For highly selective and efficient purification of mRNA for NGS applications

The GeneRead Pure mRNA Kit enables highly specific purification of mRNA ready for next-generation sequencing (NGS) applications. By effectively depleting rRNA and non-adenylated, non-coding, as well as regulatory RNA from a wide variety of eukaryotic species, the kit enriches for mRNA, ensuring optimal use of expensive sequencing capacity and resources. With a preparation time of <45 min and convenient automation options, the GeneRead Pure mRNA Kit offers unparalleled convenience by minimizing the hands-on time required to purify mRNA.

Benefits of the GeneRead Pure mRNA Kit:

- Efficient removal of almost all rRNAs (significantly >99%)
- No bias from unintended removal of poly A+ mRNAs
- As little as 100 ng–5 µg total RNA required
- Maximal convenience, minimal hands-on time
- Automation on the QIAcube®

Maximizing sequencing capacity

mRNA accounts for only 1–5% of the total cellular RNA, although the actual amount depends on the cell type and physiological state. Due to the low proportion of mRNA in the total cellular RNA pool, reducing the amount of rRNA and tRNA in a total RNA preparation increases the relative amount of mRNA. In applications such as RNA sequencing, it is of great interest to maximize the amount of information received from a sequencing run. Ribosomal RNA provides little information about the transcriptome and wastes valuable sequencing resources. The GeneRead Pure mRNA Kit effectively enriches mRNA from a wide variety of eukaryotic species, while removing rRNA and non-adenylated, non-coding, as well as regulatory RNA, assuring cost-effective use of sequencing capacity.

Exceptional convenience and cost-effectiveness

The GeneRead Pure mRNA Kit contains spin columns and all necessary reagents and buffers for the isolation of pure poly A+ mRNA from total RNA preparations in <45 minutes (Figure 1). GeneRead Pure mRNA technology combines the convenient handling of magnetic particles with the specificity of oligo-dT hybridization. The streamlined protocol only requires short pretreatment steps. ▶

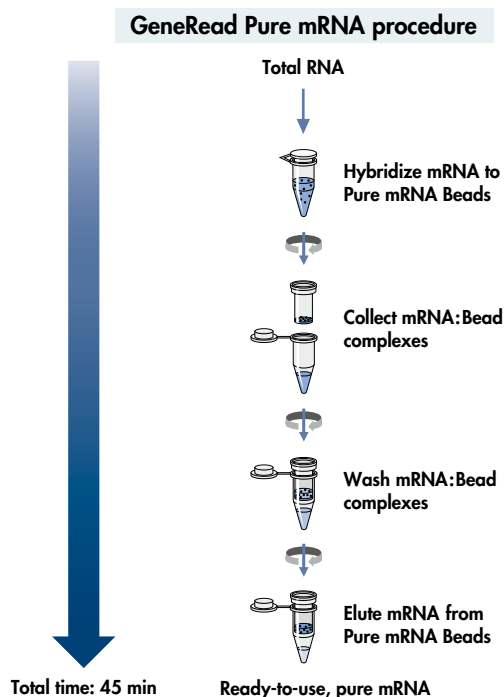


Figure 1. Convenient, streamlined procedure.



Products from other suppliers involve more time-consuming procedures as they generally require a second round of purification to achieve mRNA pure enough for NGS applications, thereby resulting in increased resources and costs. The GeneRead Pure mRNA Kit is the only kit on the market that delivers mRNA of maximal purity with only one round of purification. Poly A+ mRNA purified with GeneRead Pure mRNA technology does not require further purification and is ready for downstream use in applications such as NGS. After purification with the GeneRead Pure mRNA Kit, only meaningful reads are left — the majority of these reads are from polyA+ mRNA.

Efficient removal of ribosomal RNA

The GeneRead Pure mRNA Kit is the only mRNA purification kit to remove rRNA, including mitochondrial rRNA, almost completely (Figures 2–3 and Table 1). Due to the kit's stringent procedure, high mRNA purity is assured for increased reliability and reproducibility in sensitive downstream applications such as NGS. In addition, only small amounts of starting material, ranging from 5 µg down to 100 ng, are required.

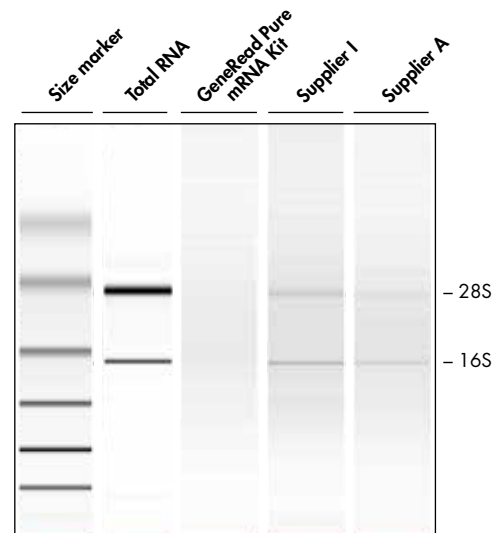


Figure 2. The GeneRead Pure mRNA Kit outperforms mRNA purification kits from other suppliers. The performance of the GeneRead Pure mRNA Kit and mRNA purification kits from other suppliers was compared. Starting material for mRNA purification was 0.5 µg total RNA. Despite only requiring one purification round, the GeneRead Pure mRNA Kit provides superior enrichment of mRNA. Residual rRNA is still visible in the form of two distinct bands in samples prepared using products from other suppliers.

Table 1. Maximal removal of rRNA

Ribosomal RNA	Removal (%)
28S	99.99
18S	99.99
5.8S	99.99
5S	99.99
16S mt	99.09
12S mt	99.78

Using the GeneRead Pure mRNA Kit, 5 µg total RNA from Jurkat cells was purified. To determine the efficacy of removal of different types of rRNA, quantitative RT-PCR was performed.

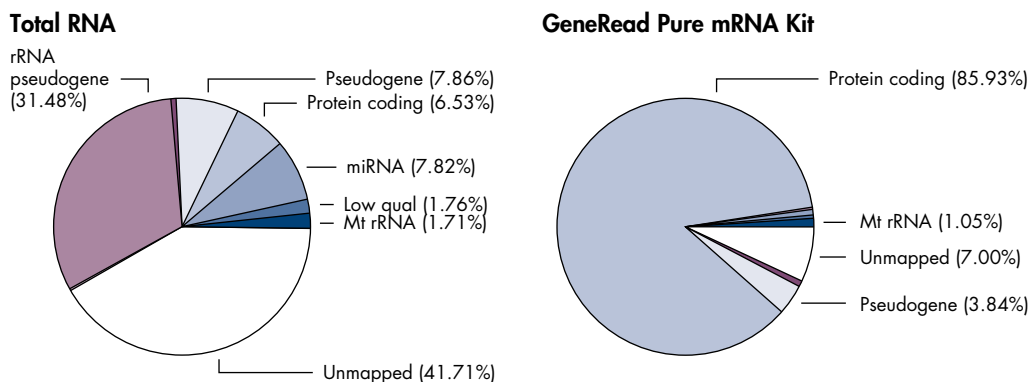


Figure 3. Specific and efficient mRNA enrichment. For analysis, 5 µg total RNA from Jurkat cells was treated using the GeneRead Pure mRNA protocol. Subsequently, libraries of the enriched samples, as well as of the total RNA were made using the same method and sequenced in multiplex by Illumina® sequencing on a MiSeq® system. Reads were mapped against the Biomart database and the percentage of mapped reads from each library for the biotypes was plotted. rRNAs are often listed under low qual. unmapped, as the annotation status of rRNA transcripts is often still suboptimal.

Minimal hands-on time required

The majority of the GeneRead Pure mRNA procedure — from hybridization and capture through to RNA cleanup — can be fully automated on the QIAcube, affording maximal convenience and greater processing flexibility.

Wide range of applications

RNA prepared using the GeneRead Pure mRNA Kit is suited for use in a wide range of downstream applications, such as:

- Next-generation sequencing
- Quantitative, real-time PCR
- Microarray analysis

Ordering Information

Product	Contents	Cat. no.
GeneRead Pure mRNA Kit (50)	For 50 reactions: Pure mRNA Beads, buffers, RNase-Free Water, Small Spin Columns, and Collection Tubes	180244
GeneRead rRNA Depletion Kit (6)	For 6 x 100 µl reactions: HMR rRNA Depletion Probes, Antibody Solution, Hybridization Buffer, BioMag Protein G Beads, RNase-free Water, RNase Inhibitor, Small Spin Columns, Reaction Tubes	180211
GeneRead rRNA Depletion Nano Kit (48)	For 48 x 100 µl reactions: HMR rRNA Depletion Probes, Antibody Solution, Hybridization Buffer, BioMag Protein G Beads, RNase-free Water, RNase Inhibitor, Small Spin Columns, Reaction Tubes	180224
GeneRead Globin mRNA Depletion Probes	Globin mRNA Depletion Probes	180950
RNeasy® Plus Universal Mini Kit (50)	For 50 RNA minipreps: RNeasy Mini Spin Columns, gDNA Eliminator Solution, Collection Tubes, RNase-Free Water and Buffers	73404
RNeasy Plus Mini Kit (50)*	For 50 minipreps: RNeasy Mini Spin Columns, gDNA Eliminator Spin Columns, Collection Tubes, RNase-Free Water and Buffers	74134
AllPrep® DNA/RNA Mini Kit (50)	For 50 minipreps: AllPrep DNA Spin Columns, RNeasy Mini Spin Columns, Collection Tubes, RNase-Free Water and Buffers	80204

* Other kit sizes available; see www.qiagen.com.

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Visit www.qiagen.com/goto/Pure-mRNA for more information!

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