

RNA quantification and analysis

Tools and Methods

The four key parameters that characterize an RNA sample

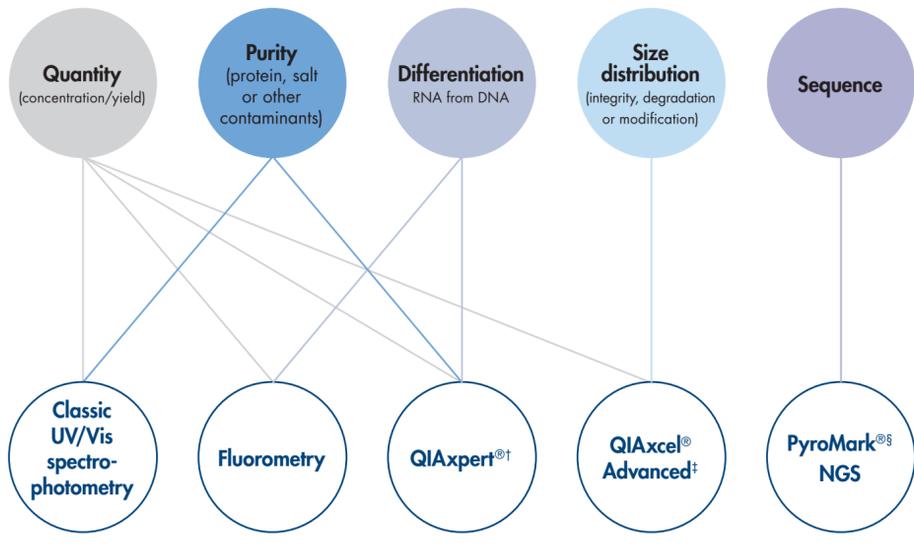
1 **Quantity**
Is there enough RNA to assay?

2 **Purity**
Is the RNA free of contaminants?

3 **Size**
Is the RNA degraded?

4 **Sequence**
Is this the right sequence?

Choosing the right technology for RNA sample assessment



* Requires a specific dye-based assay; † UV/Vis spectrophotometry combined with spectral content profiling; ‡ Fully automated capillary gel electrophoresis; § Provides quantitative sequence information

RNA quantity and purity

Concentration

RNA concentration (µg/ml) =
40 µg/ml × A₂₆₀ × dilution factor

Note: 1 OD260 Unit = 40 µg/ml for ssRNA, based on a standard 1 cm path length and neutral pH
Total RNA yield (µg) = concentration × volume of sample (ml)

Purity

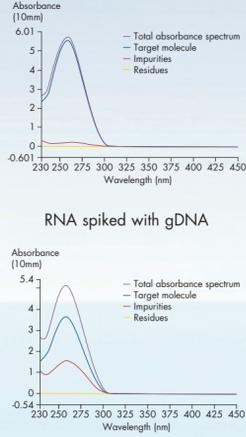
RNA purity
Pure RNA has an A₂₆₀/A₂₈₀ ratio of 1.8–2.0.
Lower value indicates protein contaminants.
Pure RNA has an A₂₆₀/A₂₃₀ ratio of >1.8.
Lower value indicates salt and other contaminants.

QIAGEN solution for nucleic acid quantification

The QIAxpert spectrophotometer provides a detailed insight into your sample's quantity and purity by analyzing your nucleic acid with spectral content profiling. This proprietary analysis feature lets you differentiate between DNA, RNA and possible sample contaminants.



- Up to 16 samples in less than 2 minutes
- 2 µl sample consumption
- Rapid analyses via touchscreen
- Easily generate reports
- USB Data output



RNA size distribution and integrity

Size of ribosomal RNAs from various sources

Bacteria	Yeast	Mouse	Human	Plant
rRNA Size (kb)				
16S 1.5	18S 2.0	18S 1.9	18S 1.9	8S 1.9
23S 2.9	26S 3.8	28S 4.7	28S 5.0	25S 3.7
				16S 1.5
				23S 2.9

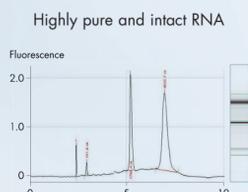
Denaturing gel analysis of total RNA. Eukaryotes: Band intensity ratio of 28S:18S rRNA should be 2:1 for intact RNA. Prokaryotes: Band intensity ratio of 23S:16S rRNA should be 2:1 for intact RNA.

QIAGEN solution for size distribution and integrity analysis

The QIAxcel Advanced uses capillary electrophoresis to automate analysis of DNA and RNA. The QIAGEN RNA Integrity Score (RIS) provides an objective quality measurement of the analyzed RNA samples and allows easy interpretation of sample integrity.



- Analyses of 12 samples in as little as 3 minutes
- Unattended analysis of up to 96 samples
- Resolution down to 3–5 bp for fragments <500 bp
- Reliable detection with sensitivity down to 0.1 ng/µl



RNA sequence

RNA sequencing (RNA-seq) is a method of investigating the transcriptome of an organism using next-generation deep-sequencing techniques. The RNA content of a sample is directly sequenced after appropriate library construction, providing an in-depth analysis of the transcriptome. Transcriptomic analyses may be validated using an independent technique, for example, quantitative PCR (qPCR).

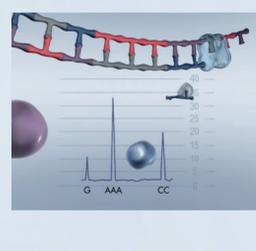


QIAGEN solution for sequence verification

Pyrosequencing is a unique detection technology that enables rapid and accurate quantification of sequence variation. The PyroMark systems are widely used for quality control and verification and validation of NGS results.



- Process 24–96 samples
- Various analysis modes
- Long and reliable read length
- Intuitive software
- Sample to result in few hours



Find out more about RNA sample analysis at www.qiagen.com/RNASampleQC.

Trademarks: QIAGEN®, Sample to Insight®, QIAxcel®, QIAxpert®, PyroMark®, Pyrosequencing® (QIAGEN Group), Agilent® (Agilent Technologies, Inc.), QIAGEN Group. Registered names, trademarks, etc. used in this document, even when not specifically marked as such, are not to be considered unprotected by law.

PROM-11757-001 SAP-1108620 © 2017 QIAGEN, all rights reserved.

Ordering www.qiagen.com/shop | Technical Support support.qiagen.com | Website www.qiagen.com