Service Profile

Targeted RNA Sequencing Service

Targeted approach for profiling low-abundance transcripts

Despite the advantages of next-generation sequencing (NGS), not every lab has the time or resources to take advantage of this technology. QIAGEN Genomic Services overcomes these limitations by combining our innovative QIAseq® RNA sequencing technology with decades of technical expertise. QIAseq Targeted RNA Panels include molecular bar code technology and provide two-stage PCR-based library preparation to deliver unbiased and accurate quantification for your targeted RNA sequencing results.

Extend your in-house resources with the expertise and service that you expect from QIAGEN. Our all-in-one Targeted RNA Sequencing Service offers the following benefits:

- **End-to-end service:** We take care of every step, from sample preparation to data analysis
- **Guidance and flexibility:** We aid in designing your project and targeted panel, and help you make the right decisions
- **Powerful digital sequencing approach:** Our optimized QIAseq technology combines unique molecular indices (UMIs) and single primer extension (SPE) to deliver unbiased and accurate quantification
- **Ready-to-publish data:** We deliver comprehensive reports and data packages, and provide guidance on the next steps

Partner with us for expert guidance and dedicated service – from Sample to Insight® – to unlock the potential of RNA sequencing in your lab today.

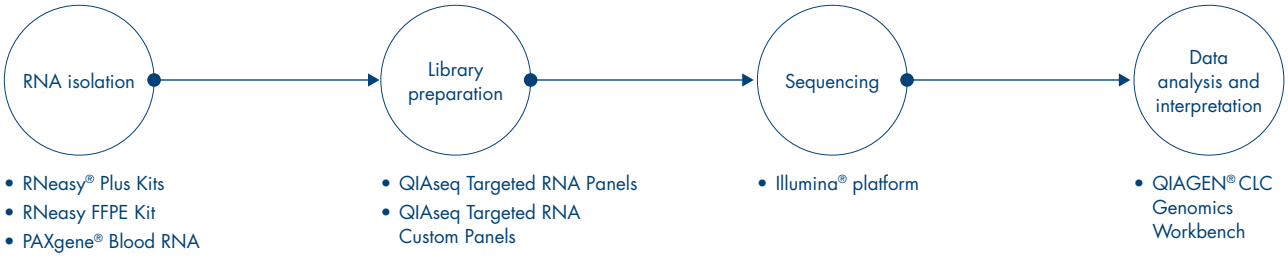

Figure 1. Sample to Insight Targeted RNA Sequencing workflow.

Advantages of targeted sequencing

Targeted sequencing is particularly advantageous for achieving high coverage of regions or genes of interest while keeping the cost of sequencing and complexity of data interpretation manageable. In RNA analysis, a targeted approach can provide more evidence of low-abundance transcripts. Existing target enrichment, library preparation and sequencing steps are based on DNA polymerase and amplification processes, introducing substantial bias and artifacts. QIAseq Targeted RNA Panels unique molecular barcode (or molecular tag, MT) technology into a highly multiplexed, PCR-based target enrichment process, enabling unbiased and accurate quantification of a targeted panel of mRNA transcripts using NGS.

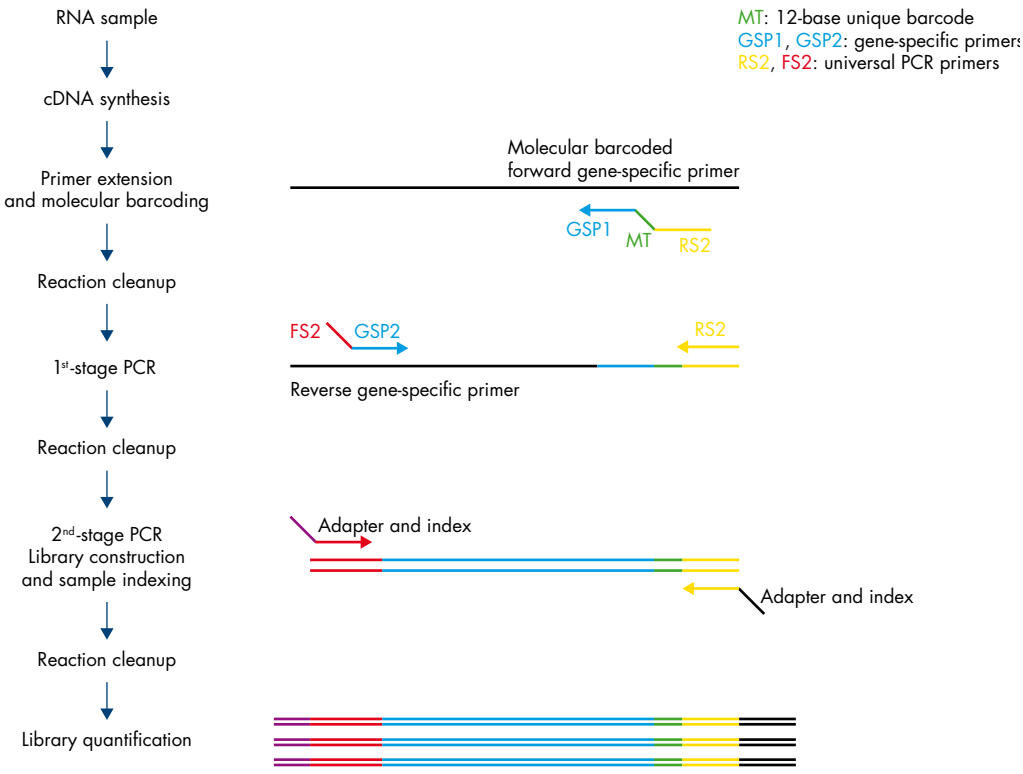

Figure 1. Uniquely tagged cDNA. Starting with 25 ng of total unfragmented RNA, cDNA is synthesized, and each cDNA molecule is tagged with a unique molecular bar code (molecular tag, MT) before any amplification. The uniquely tagged cDNA molecules then undergo a two-stage PCR step for enrichment and library construction.

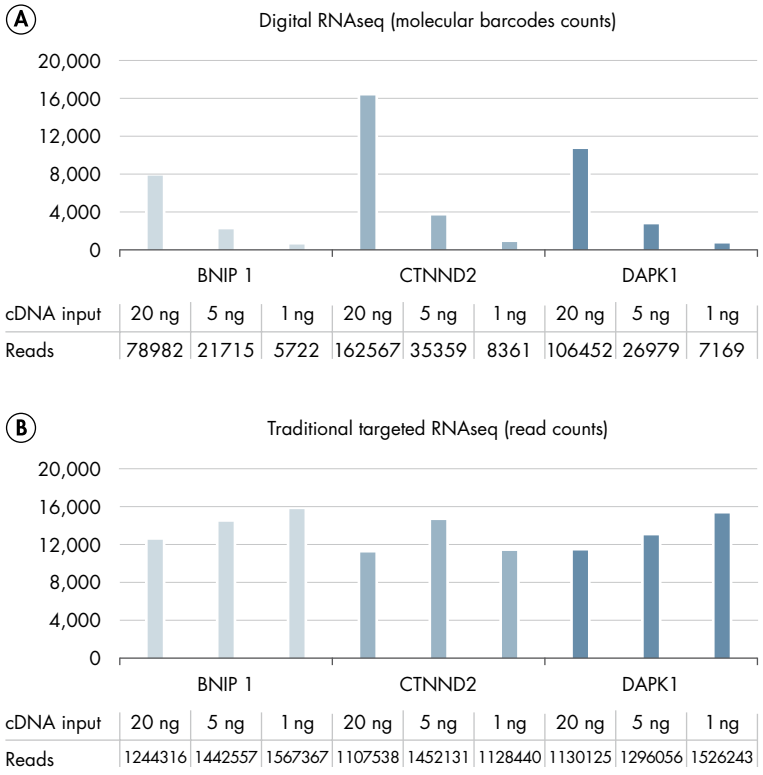

Figure 2. Unbiased and accurate gene quantification. Different input amounts (20 ng, 5 ng or 1.25 ng) of universal reference RNA were used to determine expression levels of three genes (BNIP1, CTNND2 and DAPK1) using targeted RNA-seq. QIAseq targeted RNA sequencing method (A; UMI counts) showed accurate quantification of all three genes corresponding to different RNA input, whereas traditional targeted RNA-seq (B; read counts) revealed PCR duplication limitation and yielded inaccurate quantification.

Service specifications

Consultation	Free consultation with an expert to design an experimental setup that best meets your needs.		
Sample requirements	**Sample input**	**Isolation kit**	**Input requirements**
	Customer-isolated RNA	N/A	Minimum: 50 ng Recommended: 450 ng >5 ng/µl OD_{260}/OD_{280} value: >1.6 RIN value: >7
	Cells	RNeasy Plus Kit	Minimum: 1 x 10^6 cells pelleted and frozen
	Fresh frozen tissue	RNeasy Plus Universal Kit	Minimum: 4–5 mg Maximum: 50 mg
	FFPE	RNeasy FFPE Kit	Minimum: 2 x 10 µm sections of 250 mm^2 Maximum: 4 x 10 µm sections of 250 mm^2
	Blood (PAXgene)	PAXgene Blood RNA	Recommended: 1 tube
	Other	**Please inquire**	
Sample quality control	Fluorescence-based dye for determination of sample concentration. Gel electrophoresis for determination of RNA integrity (e.g., RIN value from capillary gel electrophoresis or DV_{200} for FFPE RNA). This is a STOP/GO point where it is possible to omit samples or replace samples before proceeding.		
Library preparation and quality control	Library preparation using QIAseq Targeted RNA Panel or custom panel. Library QC by gel electrophoresis to check for the right fragment size and concentration. Integrated UMI technology.		
	This is a STOP/GO point where it is possible to omit samples before proceeding.		
Sequencing parameters	Illumina NextSeq®, MiSeq® or NovaSeq® systems Single-end reads Read length of 150 bp The recommended read depth depends on the sample amount and quality as well as the panel.		
Complete data analysis	Raw data	De-multiplexed FASTQ files	
	Raw data quality control	CLC graphical quality control report (for each sample) CLC supplementary quality control report (for each sample)	
	Data trimming	CLC trim report (for each sample) (removal of adapters, low-quality, short sequences and ambiguous nucleotides)	
	Mapping (GRCh38 or GRCm38) and quantification	Counts matrix (including read count, UMI count, mean reads per UMI, total gene reads, expression value, TPM normalized counts)	
	Supported species	Homo sapiens, Mus musculus	
	Merge data with data from previous projects	**Inquire**	
	Data delivery	Encrypted USB/HDD or cloud delivery	

Final report and consultation	The final data analysis package contains an overview of data analysis and algorithms used, the files and tables listed above and publication-ready figures (PDFs provided as standard; inquire for SVG or other formats). A teleconference is scheduled with QIAGEN scientists to discuss analysis and validation of results. Consultation and support will be provided for 90 days following delivery of data (for data delivery only projects), or delivery of data analysis (for data analysis inclusive projects). **Inquire** for extended support beyond 90 days.

Note: Service specifications might be tailored to the needs of the project on a case-by-case basis.

How can we accelerate your research?

Our expert team is looking forward to learning about your research project and designing your customized service with QIAGEN.

Tell us about your project

The QIAGEN Genomics Service is intended exclusively for research use only (RUO). This service is not intended for the diagnosis, prevention or treatment of a disease.

For up-to-date licensing information and product-specific disclaimers, see the respective QIAGEN kit handbook or user manual. QIAGEN kit handbooks and user manuals are available at **www.qiagen.com** or can be requested from QIAGEN Technical Services or your local distributor.

1124690 05/2022