GENEWIZ MULTIOMICS & SYNTHESIS SOLUTIONS | NEXT GENERATION SEQUENCING

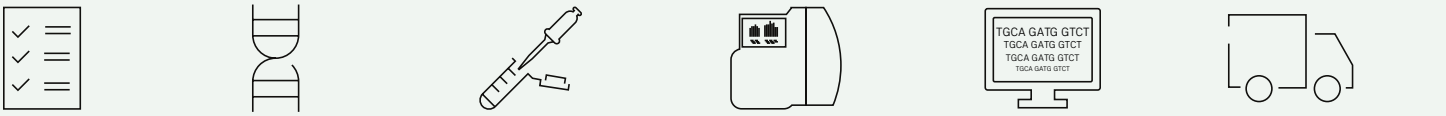
RNA-Seq Technical Specifications

RNA Sequencing Services

- Standard RNA-Seq
- Strand-Specific RNA-Seq
- Small RNA-Seq
- Ultra-Low Input RNA-Seq
- Single-Cell RNA-Seq*
- Long-Read RNA-Seq using PacBio or Oxford Nanopore Technologies*
- Spatial Transcriptomics*
- CLIA RNA-Seq*

*See **genewiz.com** for technical specifications.

RNA Sequencing Workflow



1 Experimental Design
Define objectives and design assay accordingly

2 Sample Preparation
Extract and purify input RNA

3 Library Preparation
Enrichment, cDNA synthesis, and adapter ligation

4 Sequencing
Sequence libraries using NGS platforms

5 Data Analysis
Trim, filter, and map reads; perform advanced analysis

6 Project Delivery
Receive data files and post-delivery support

1 Experimental Design

We offer resources to help you find the best NGS solution and experimental design for your project.

Interactive NGS Solution Selection Tool:
genewiz.com/ngs

Contact us for a **free technical consultation** with a Ph.D.-level scientist
web.genewiz.com/ngs-inquiry

*Other sample types accepted. View Sample Submission Guidelines for details.
†Please inquire about submitting lower inputs.
‡Contact us about our RNA Stabilization Tubes to ship RNA samples at ambient temperature.

2 Sample Preparation

Our extraction solutions are compatible with over 30 standard and hundreds of custom sample types to accommodate your starting material. The below table is by no means an exhaustive list.

Sample Type*	Minimum Amount†	Recommended Amount
Total RNA‡	Standard: 150 ng, 25 µL, ≥6 ng/µL Ultra-Low: ≥50 pg, 3-5 µL, ≥10 pg/µL	Standard: ≥500 ng, 25-50 µL, 20-50 ng/µL Ultra-Low: ≥10ng, 3-5µL, ≥2ng/µL
Eukaryotic cell pellet	1×10^4 cells	≥1×10^6 cells
Prokaryotic cell pellet	1×10^6 cells	≥1×10^8 cells
Frozen tissue	2 mg	30 mg
FFPE	2 curls, 5µm-20 µm, >150 mm^2	≥4 curls, 5µm-20 µm, >150 mm^2

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3 Library Preparation

RNA-Seq Service	Target RNA	RNA Selection Method
Standard & Strand-Specific	mRNA (eukaryotic)	Poly(A) selection
	mRNA + lncRNA	rRNA depletion
Small	Small RNA (miRNA, siRNA, piRNA)	Size fractionation with adapter ligation to 5' phosphate
Ultra-Low Input	mRNA (eukaryotic)	Poly(A) selection with enrichment for full-length transcripts

4 Sequencing

Platform	Illumina® NovaSeq™
Configuration	2x150 bp
Depth	Customizable to your project needs*
Data Quality	Guaranteed ≥85% bases with Q30 or higher

*Generally, we recommend 5-10 million read pairs per sample for small genomes (e.g. bacteria) and 20-30 million read pairs per sample for large genomes (e.g. human, mouse). Medium genomes often depend on the project, but 15-20 million read pairs per sample is typically sufficient. For *de novo* transcriptome assembly projects, we recommend 100 million read pairs per sample.

5 Data Analysis

RNA-Seq Service	Standard Analysis Package	Additional Analysis Options
Standard Strand-Specific Ultra-Low Input	• Trimming • Mapping • Differential gene expression	• Gene fusion discovery • RNA SNP/INDEL detection • Novel transcript discovery • *De novo* transcriptome assembly
Small	• Trimming • Mapping • Differential gene expression • Small RNA discovery	

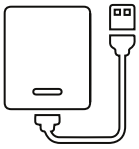
6 Project Delivery

Deliverables for All Projects	Optional Deliverables
• **Sample quality control report** • **Raw data (FASTQ files)**	• Aligned data (BAM file) • Hit counts (TXT file) • DGE results (CSV file) • GO enrichment analysis (CSV file) • Differential splicing analysis (DEXSeq report) • De-multiplexed, aggregated Picard BAM file with summary metrics

DATA DELIVERY OPTIONS

SFTP

Customer Cloud Account



External Hard Drive

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