# RNA-Seq Technical Specifications



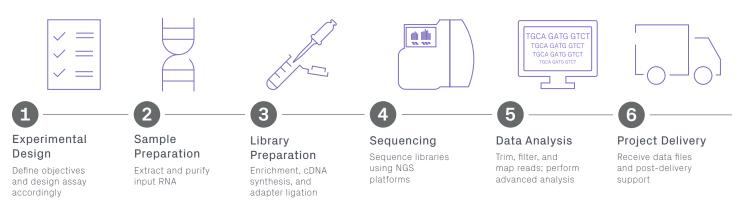
## Azenta Life Sciences RNA Sequencing Services

- Standard RNA-Seq
- Strand-Specific RNA-Seq
- Small RNA-Seq
- Ultra-Low Input RNA-Seq

- Single-Cell RNA-Seq\*
- Iso-Seq\*
- Digital Spatial Profiling\*

\*Not covered here. See genewiz.com for more details.

## **RNA Sequencing Workflow**



### Experimental Design

Azenta Life Sciences provides 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

# 2 Sample Preparation

Sample Type*	Minimum Amount <sup>†</sup>	Recommended Amount
Total RNA <sup>‡</sup>	500 ng (standard) 10 pg (ultra-low)	2 µg
Eukaryotic cell pellet	10 <sup>4</sup> cells (standard) 1 cell (ultra-low)	10 <sup>6</sup> cells
Prokaryotic cell pellet	10 <sup>6</sup> cells	10 <sup>8</sup> cells
Frozen tissue	2 mg	10 mg
FFPE	2 slides	4 slides

Other sample types accepted. View <u>Sample Submission Guidelines</u> for details. \*Please inquire about submitting lower inputs.

<sup>‡</sup>Contact us about Azenta Life Sciences' RNA Stabilization Tubes to ship RNA samples at ambient temperature.



# **RNA-Seq Technical Specifications**



Library Preparation

RNA-Seq Service	Target RNA	RNA Selection Method
Standard & Strand-Specific	mRNA (eukaryotic)	Poly(A) selection
	mRNA + IncRNA	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

### Sequencing

Platform	Illumina® NovaSeq™ or HiSeq®	
Configuration	2×150 bp	
Depth	Customizable to your project needs*	
Data Quality	Guaranteed ≥80% 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.

Data Delivery Options



SFTP ٦Ôp SFTP

### **Project Delivery**



Customer Cloud Account



External Hard Drive (US Only)



## **Data Analysis**

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

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



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