Service Profile

mRNA Sequencing Service

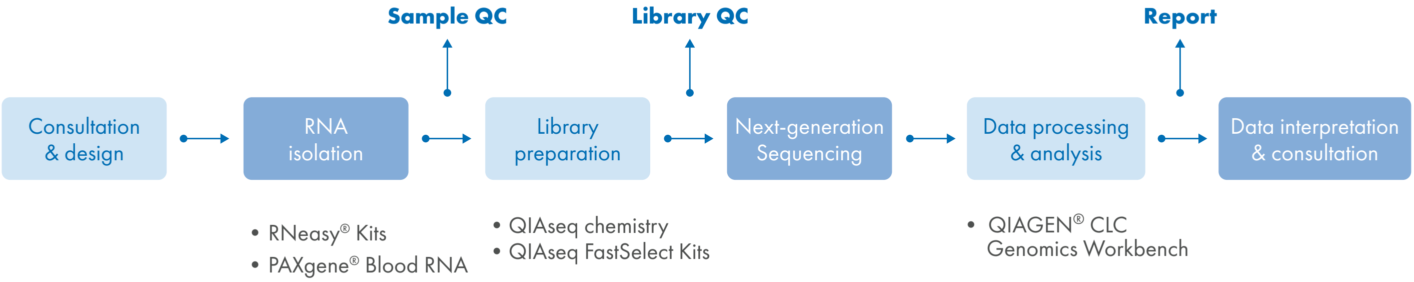
Every step of your mRNA sequencing, quality control and analysis is designed for success

Our mRNA Sequencing Service enriches for poly-A tailed transcripts or depletes rRNA with QIAseq FastSelect to increase the sequencing depth for coding mRNAs, which improves the sensitivity to mRNAs expressed at low levels. In addition, the library preparation retains information about which of the two DNA strands was used to transcribe a given RNA. This information provides increased confidence in transcript annotation and enables detection of antisense transcript expression. Further, during data analysis , highly abundant mitochondrial poly A transcripts can be filtered out to further enhance the resolution of the target molecule.

mRNA sequencing is recommended for discovery work and especially for differential expression analysis. Paired-end sequencing increases the mapping percentage to poorly annotated genomes and makes it possible to identify splice variants with much higher confidence.

- **End-to-end service:** We take care of every step, from sample preparation to data analysis
- **Full-spectrum solution:** We provide a seamless flow from biomarker discovery to clinical assay development and approval
- **Insightful data analysis:** Pathway, upstream regulators and disease analysis of differentially expressed genes can be provided with the industry-leading Ingenuity Pathway Analysis software

Partner with us for expert guidance and dedicated service – from Sample to Insight® – for profiling your samples today.



Bioinformatics: a bridge between data and discovery

Novel insights often remain elusive without the right tools and expertise for data analysis and interpretation. QIAGEN Genomic Services use industry-leading pipelines and best-in-class algorithms to provide you with the answers to your biological questions. Below are some examples of data analysis results, including publicationgrade graphs and figures, which are part of the Genomic Services deliverables.

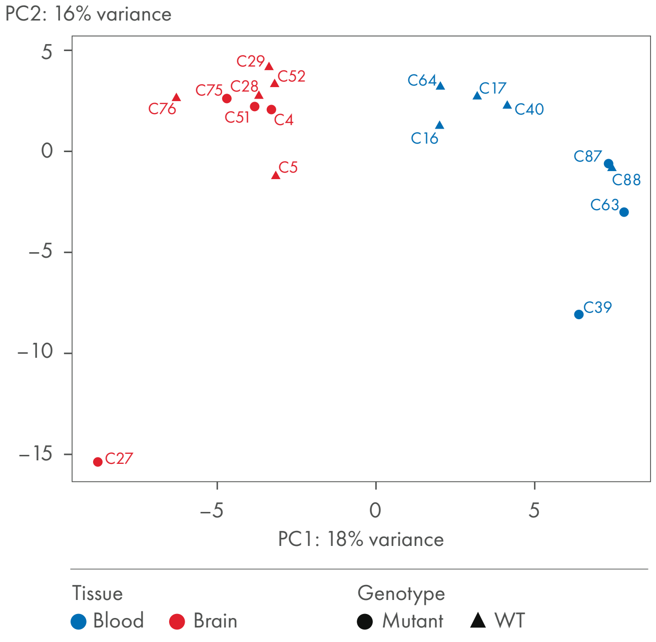


Figure 2. Principal component analysis (PCA) plot of gene expression profiles for three biological replicates belonging to four different experimental groups (codified by different colors). PCA is applied to variance-stabilizing transformation of row gene counts, and each sample is displayed as a dot in the first two principal components spaces (PC1 and PC2). The PCA plot allows display and visually assess of overall dissimilarity among samples and experimental groups.

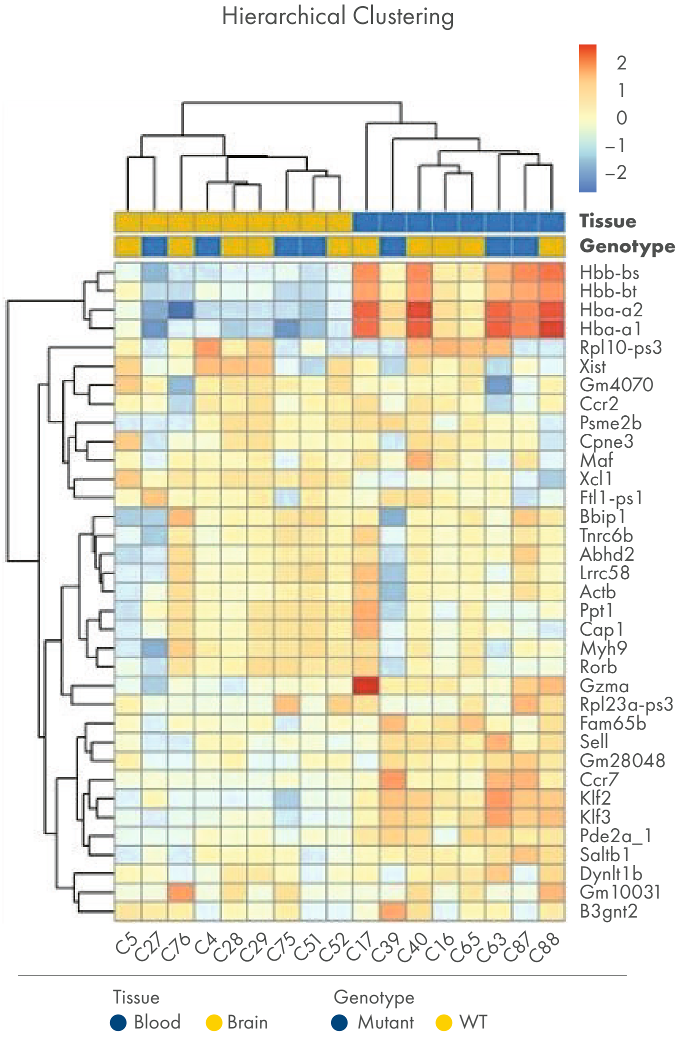


Figure 3. Heatmap illustrating the clustering analysis performed on the variance stabilizing transformed counts of the top 35 highest variance genes. Each row represents a specific gene, columns indicate samples, and color codifies difference of the expression level to the row mean.

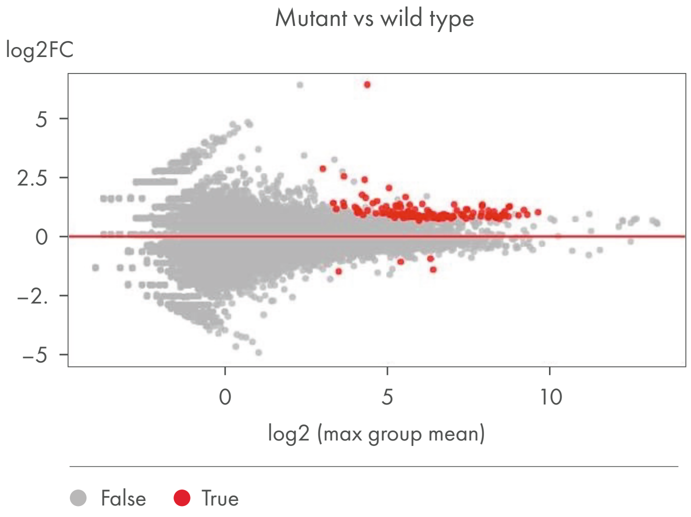


Figure 4. For each differential gene expression analysis, the result of the statistical test is represented in a separate MA plot. Each gene's fold-change is plotted against its mean expression among all samples. All significantly differentially expressed genes are marked in red. Significant changes are defined as FDR<0.01.

Service specifications

Consultation

Free consultation with an expert to design an experimental setup that best meets your needs.

Sample requirements and extraction

Sample input	Extraction kit	Input requirements
Customer-isolated RNA	N/A	Recommended: at least 200 ng total RNA (>5 ng/µl)
Cells	RNeasy Plus	Minimum: 2 x 10^6 cells, pelleted and frozen Maximum: 1 x 10^7 cells pelleted and frozen
Fresh frozen tissue	RNeasy Plus Universal	Minimum: 5 mg Maximum: 50 mg
FFPE	RNeasy FFPE	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	**Inquire**	

Sample quality control

Fluorescence-based dye for determination of sample concentration

Electrophoresis for determination of RNA integrity (e.g., RIN value from capillary gel electrophoresis)

This is a STOP/GO point with appropriate assessment and suggestions in sample replacement or sample selection as needed.

Library preparation and quality control

Library preparation using QIAseq chemistry

Library quality control by capillary gel electrophoresis to check for the right fragment size and concentration

This is a STOP/GO point with appropriate assessment and suggestions in sample selection as needed.

Sequencing parameters

Single-end or paired-end reads

Read length of 75 bp

Read depth of 1 x 30 M reads or 2 x 25 M to 30 M reads, on average

Service specifications

Complete data analysis and report	Final data analysis package contains the following: • Overview of materials and methods, data analysis and algorithms used summarized in an HTML report • Files and tables as described below • Inquiry for specific publication-ready figures (PDF, SVG or other formats)		
	Project report	Overall project report that includes information about sequencing and data analysis of the project as well as materials and methods	HTML
	Metadata table	Table detailing all the sample associated information (including all relevant description provided by the customer)	Excel
	Raw data	De-multiplexed compressed FASTQ files	FASTQ.GZ
	Raw data quality control	CLC graphical QC report (for each sample)	PDF
		CLC supplementary QC report (for each sample)	Excel
	Data trimming	CLC trim report (for each sample) Removal of adapters, low-quality, short sequences and ambiguous nucleotides	PDF
	Mapping	RNA-seq report (for each sample and combined) Read count statistics or mapping rates, fragment counts, distribution of biotypes, transcript coverage	PDF
	Quantification	Raw counts matrix	Excel
		TPM-normalized counts matrix	Excel
	Unsupervised analysis	PCA plot	Included in the HTML report
		Hierarchical clustering heatmap	Included in the HTML report
	Differential expression for each defined comparison (maximum 10) **Inquire** for additional comparison/analysis	Differential expression statistics Fold-change, log2 FC, p-value, FDR-corrected p-value, Bonneferoni corrected values	Excel
		MA plot for each differential expression comparison	Included in the HTML report
	Pathway analysis	QIAGEN Ingenuity Pathway Analysis® (IPA®) Available as an add-on; refer to IPA demo report. Supported for human, rat and mouse. **Inquire** for other species.	Various (Excel, PDF)
	Species supported	*Bos Taurus, Caenorhabditis elegans, Canis familiaris, Danio rerio, Drosophilia melanogaster, Equus caballus, Gallus gallus, Homo sapiens, Mus musculus, Oryza satvia, Pan troglodytes, Rattus norvegicus, Sus scrofa.* **Inquire** for other species.	
	Merge data with data from previous projects	**Inquire**	
	Data delivery	Cloud delivery or USB/HDD (encrypted USB/HDD inquire)	
Consultation	Teleconference with QIAGEN scientists to discuss analysis and validation of results using miRCURY qPCR assays. 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.		
	Data storage	Your data will be retained on the Genomic Service servers for 90 days from the date of data delivery after which point your data will be deleted from our servers. Please ensure your data is downloaded by this time-point. **Inquire** for extended storage 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 discovery?

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

Accelerate your discovery

One partner for all your needs

Expertise at your fingertips

Insights powered by science

Tell us about your project

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