mRNA Sequencing

mRNA sequencing (mRNA-Seq), a method of transcriptome profiling based on deep- sequencing technologies, providing a more precise measurement of transcript levels and their isoforms with a complete snapshot of the coding transcriptome than other methods such as microarray analysis.

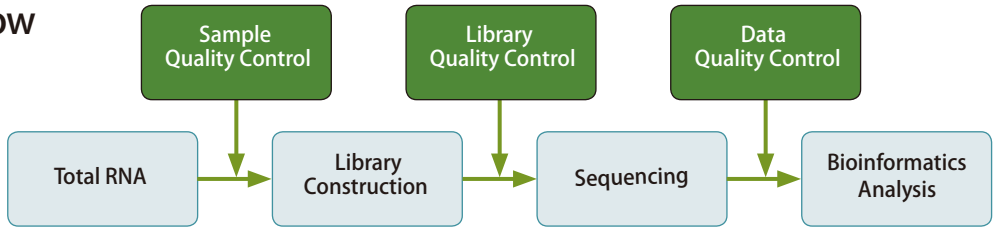
Transcriptome sequencing with longer read length enables the identification of novel transcripts, alternative splicing and gene fusions, and scientists can identify biomarkers or key regulated genes, key functional genes related to different phenotypes, as well as key factors in temporal changes. RNA-Seq quantification with shorter read length can simultaneously measure the expression levels of many transcripts. It is widely used in disease research, drug response research, pharmacokinetics, and personalized healthcare research.

Novogene offers comprehensive transcriptome sequencing and RNA-Seq quantification services using cutting-edge Illumina platform with different sequencing strategies and widely accepted analysis software, serving customers with different research emphases.

The Novogene Advantage

- Extensive experience with >30,000 samples successfully sequenced in over 3,000 completed projects and articles published on transcriptome sequencing results across different species.
- Comprehensive data analysis using widely accepted mainstream software and mature in-house pipeline to discover novel transcripts, differential expressions, and function annotations.
- Unsurpassed data quality with a guaranteed Q30 score ⩾ 80% that exceeds Illumina's official guarantee.

Project Workflow



SEQUENCING STRATEGY

- 250~300 bp insert cDNA library
- Illumina platform, paired-end 150 bp (Transcriptome sequencing)

RECOMMENDED SEQUENCING DEPTH

- Minimum:
  6G for animals and plants
  2G for fungi
- Recommended:
  15G for animals and plants
  3G for fungi

DATA QUALITY GUARANTEE

- Our data quality guarantee, as measured by the percentage of bases with a sequencing quality score above Q30 (PE150, ⩾ 80%), exceeds Illumina's official guarantee (PE150, ⩾ 75%)

TURNAROUND TIME

- Within 15 working days from verification of sample quality without data analysis.
- The turnaround for data analysis is project dependent.

SAMPLE REQUIREMENTS

- Total RNA amount: ⩾ 0.8 μg; (For human and mouse, 50ng could be accepted with low-input method; for animal and plant, 100ng could be accepted with low-input method)
  RNA concentration: ⩾ 50 ng/μl
- RIN value ⩾ 6.3 for plants and fungi; RIN value ⩾ 6.8 for animals
- OD260/280 ⩾ 2.0, OD260/230 ⩾ 2.0, without degradation and DNA contamination
- FFPE sample: > 10 scrolls or slides. Samples should be tested as pre-qualified by gel electrophoresis before sample submission.

Representative data quality results of mRNA sequencing (PE 150) from Novogene

Sample Name	# Of Raw Reads	# Of Clean Reads	Clean Bases	Error Rate (%)	Q20 (%)	Q30 (%)	GC Content (%)
a	61857270	58272366	8.74G	0.02	96.55	91.82	49.63
b	63461444	59634406	8.95G	0.02	96.66	91.97	47.74
c	54854516	51178674	7.68G	0.02	96.46	91.56	49.55
d	54204202	52191586	7.83G	0.02	96.46	91.53	51.54
e	62043078	59714752	8.96G	0.02	96.31	91.18	51.27
f	76030470	72813412	10.92G	0.02	96.38	91.40	53.36

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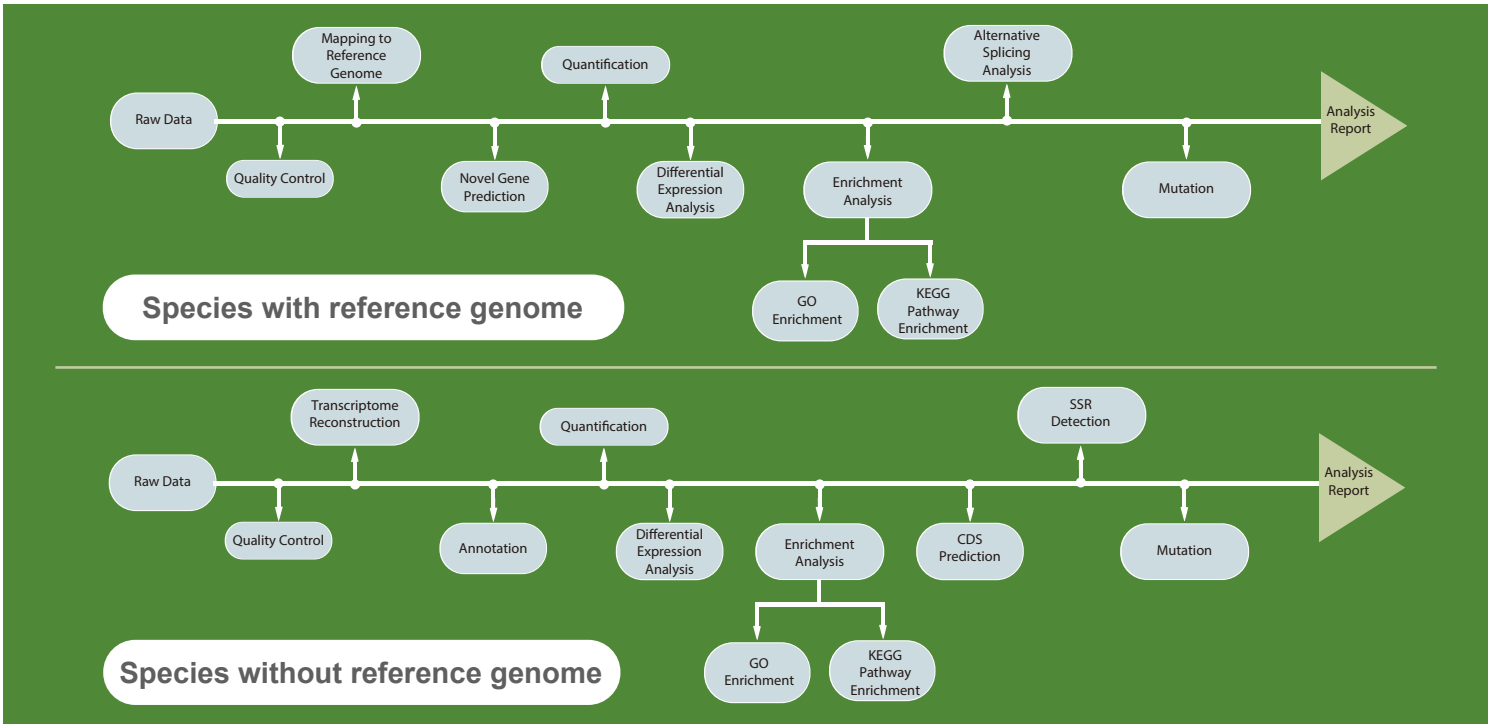
Analysis Pipeline



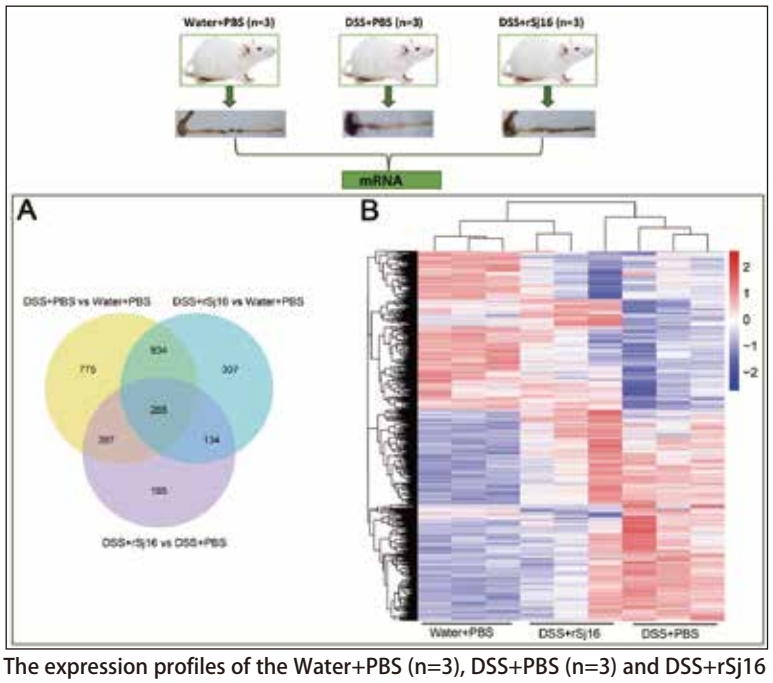
Project Example

The following study utilized Novogene's expert mRNA-Seq & lncRNA & miRNA services.

rSj16 Protects against DSS-Induced Colitis by Inhibiting the PPAR-α Signaling Pathway

Theranostics, 2017, 7(14):3446-3460

As published online in Theranostics, the preliminary analyzes of LncRNA & mRNA& miRNA were performed by Novogene, reported that lncRnA, mRNA and small RNA transcriptomes provide insights into the protective effects of rSj16 on DSS-induced colitis, which is mainly mediated by PPAR-α signaling pathway inhibition. rSj16 attenuated clinical activity of DSS-induced colitis mice, diminished pro-inflammatory cytokine production, up-regulated immunoregulatory cytokine production and increased Treg percentages in DSS-induced colitis mice. Moreover, DSS-induced colitis mice treated with rSj16 displayed changes in the expression levels of specific genes in the colon and show the crucial role of peroxisome proliferator activated receptor α (PPAR-α) signaling pathway. PPAR-α activation diminished the therapeuticeffects of rSj16 in DSS-induced colitis mice, indicating that the PPAR-α signaling pathway plays a crucial role in DSS-induced colitis development.The findings of this study suggest that rSj16 may be useful as a therapeutic agent and that PPAR-α may be a new therapeutic target in the treatment of IBD.



The expression profiles of the Water+PBS (n=3), DSS+PBS (n=3) and DSS+rSj16 (n=3) groups. (A) Venn diagram showing the genes that were unique to each group and shared among the three groups. (B) The genes that were differentially expressed among the groups are shown as heatmaps

EXAMPLES OF PUBLICATIONS USING NOVOGENE'S EXPERTISE

Year	Journal	Article
2018	Genome Biology	The RNA Binding Protein SORBS2 Suppresses Metastatic Colonization of Ovarian Cancer by Stabilizing Tumor-Suppressive Immunomodulatory Transcripts
2018	Nucleic Acids Research	Pold3 is Required for Genomic Stability and Telomere Integrity in Embryonic Stem Cells and Meiosis
2017	Neuron	A Critical Role of Presynaptic Cadherin/Catenin/p140Cap Complexes in Stabilizing Spines and Functional Synapses in the
2016	Cell Stem Cell	SIRT6 Controls Hematopoietic Stem Cell Homeostasis through Epigenetic Regulation of Wnt Signaling
2016	Genome Research	Genome-Wide A-to-I RNA Editing in Fungi Independent of ADAR Enzymes
2016	Genome Research	RNA-seq of 272 Gliomas Revealed a Novel, Recurrent PTPRZ1-MET Fusion Transcript in Secondary Glioblastomas
2016	Nature Structural & Molecular Biology	Single-cell RNA-Seq Profiling of Human Preimplantation Embryos and Embryonic Stem Cells
2016	Nature Communications	Integration of Hippo Signalling and the Unfolded Protein Response to Restrain Liver Overgrowth and Tumorigenesis