

# RNA-Seq or nCounter®?

## Key Considerations for Gene Expression Studies

Advances in gene expression profiling have enabled researchers to obtain gene expression data from virtually any sample type – down to the single-cell level. At Canopy Biosciences, we recognize that every gene expression study is unique and requires a tailored approach to measure transcript expression. We offer both RNA sequencing and NanoString® nCounter® assays to provide the speed, breadth, and sensitivity our customers may require. Here, we present key questions to determine the best approach for your research.

### 1. Is your study discovery-based or hypothesis-driven?

RNA-Seq enables unbiased quantification of virtually any transcript from a wide range of sample types. By capturing both known and novel features, RNA-Seq best supports discovery-based research. The nCounter® platform from NanoString® offers a more targeted-approach to measuring transcripts to support hypothesis-driven studies. Select from one of many pre-designed nCounter® panels to quantify 800+ targets per run. If your panel is missing genes critical to your research, Canopy can easily customize a panel with up to 30 additional targets.

### 2. How quickly do you need results?

Fast turnaround times are one of the greatest advantages of nCounter® assays. Due to a simple wet-lab workflow and rapid data acquisition, Canopy Biosciences can deliver data for projects with up to 48 samples in under 2 weeks. Because the volume of data collected is generally much greater, the standard turnaround time for RNA-Seq assays starts at 4-6 weeks.

### 3. Do you have bioinformatics support?

Analyzing RNA-Seq data from FASTQ files requires knowledge of programming languages or access to bioinformatics pipelines. nCounter® data is far simpler and expressed in terms of raw counts normalized to housekeeping genes. The instrument's RCC files are small and can be easily transferred through email, while FASTQ files must be transferred through the cloud.

Canopy offers analysis for both nCounter® and RNA-Seq datasets through ROSALIND – an interactive analysis interface for differential gene expression that includes heat maps, volcano plots, and pathway analysis. Let us provide an intuitive data visualization space that makes it easy to collaborate with colleagues to create publication-ready figures.



#### 4. What type of samples are you working with?

Formalin-fixed paraffin-embedded (FFPE) samples are notoriously challenging for molecular analysis due to high variability, low yield, and high degradation. Canopy scientists have spent years optimizing extraction protocols to yield high quality RNA from a variety of sample types – including FFPE samples. Despite technical expertise, working with FFPE samples remains a challenge.

nCounter® employs molecular barcodes to target genes with great sensitivity without the need for any amplification steps that may introduce bias. The result is improved detection of low-expression RNA from smaller amounts of starting material. Despite the many advantages of RNA-Seq, sources of sequencing bias and systematic noise make RNA-Seq vulnerable to the general biases and errors of most next-generation sequencing technologies. One side-effect is that a larger amount and higher quality starting material is required to achieve appropriate coverage.

#### 5. Are you studying gene fusions or transcript isoforms?

Accurate detection of gene fusions is essential for diagnostic and research purposes. Due to detection constraints, many platforms have low throughput when quantifying these rearrangements. RNA-Seq overcomes these limitations by providing genome-wide detection as well as discovery of novel fusion genes. nCounter® can also be used to sensitively detect known fusion transcripts but is limited to the specific fusion partners included in the panel design.

#### 6. Do you need a CLIA-certified laboratory?

Canopy Biosciences is uniquely positioned to support experiments to develop laboratory developed tests (LDT) or companion diagnostic (CDx). Our RNA-Seq services provide a global

snapshot of transcriptional changes which can then lead to the selection of a reduced set of genes to stratify patient populations. nCounter® assays can be expediently run in our CLIA-certified laboratory to provide real-time data during ongoing clinical trials. RNA-Seq and nCounter® are truly complimentary platforms and the findings from one experiment may be used to support the design of the next. Streamline your next project by working with our experienced laboratory.

*To learn more, visit us at [canopybiosciences.com](https://canopybiosciences.com)*

#### References

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