

Total RNA Sequencing

Incorporating globin and ribosomal depletion

Product Overview

Total RNA sequencing is an invaluable research tool that provides a more holistic view of the transcriptome. This can include insight into both coding and non-coding regions with roles in regulatory mechanisms and the detection of alternative splicing events and post-transcriptional modifications that provide a deeper understanding of gene regulation and isoform diversity.

Broad Clinical Labs (BCL) has performed a thorough evaluation of available technologies and processing conditions to provide the highest quality total RNA (with a combined globin depletion / rRNA depletion step) by working closely with scientists within and outside the Broad Institute. The BCL generates cDNA libraries that are depleted of globin and rRNA regions and possess a superior quality with increased complexity. This increased complexity enables us to achieve a high sequencing depth of up to 100 million reads aligned in pairs while keeping the duplication rates low. The enhanced complexity of these libraries facilitates the identification of a greater number of transcripts and protein-coding genes compared to our conventional mRNA workflow (Fig 1).

BCL is able to offer a highly scalable total RNA product with rRNA and globin depletion by leveraging core competencies in process design, molecular biology, laboratory automation, and integrated LIMS and analysis tools. RNA samples are processed using a globin/rRNA depletion and stranded cDNA library construction sample preparation kit modified to include custom UMI adapters and primers for improved performance, multiplexing, and integration into our automated platform. RNA library quantity is verified by both PicoGreen™ QC and qPCR prior to sequencing.

Total RNA Seq with Depletion (50M reads aligned in pairs) starting at \$200

Pricing dependent on number of samples and project details

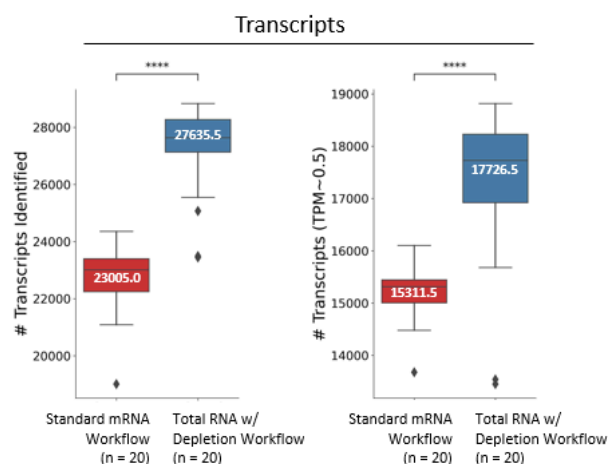


Figure 1. Comparison of the number of transcripts detected in 20 blood samples processed with both mRNA and Total RNA workflows.

What's Included

- Sample receipt
- Combined globin RNA and rRNA depletion, stranded cDNA synthesis, and library construction with UMIs
- Illumina Sequencing (2x 151bp Reads) on NovaSeqX

Input Requirements

- PaxGene Whole blood, or purified RNA derived from whole blood or fresh frozen tissue
- ≥ 500ng total in 50-300uL volume (30ng/uL concentration is recommended) of purified total RNA in nuclease-free water
- Minimum 24 samples/batch
- A260/230 >1.3
- RIN > 3.5 required
- Minimum sample data including collaborator participant or sample ID, sample tube barcode, sample type

Data Deliverable

- 50, 100, and 200 Million reads aligned in pairs.
- CRAM file aligned to human genome assembly (hg38)