Navigating Next-Generation Pre-Clinical Models of Drug Discovery & Development with RNA-seq

ATCC 100 YEARS

Abstract #1068

Ajeet P. Singh, PhD; Rula R. Khairi, MS; Amy L. Reese, MS; Noah Wax, MS; Steve King, MS; Jade L. Kirkland, MS; James Duncan, BS; Robert Marlow, BS; Ana Fernandes, BS; Jonathan L. Jacobs PhD

ATCC, Manassas, VA 20110

Abstract

Cell models paired with curated gene expression data are vital for preclinical research. Transcriptomic data from traceable, authenticated cell lines streamline hypothesis development and experimental design while illuminating underlying biological mechanisms. In cancer research, these models have notably identified therapeutic targets and biomarkers. To advance cancer research, ATCC® sequenced the transcriptomes of 141 immune cancer cell lines, accessible through ATCC Cell Line Land, encompassing various hematopoietic lineages. This effort provided insights into gene expression patterns, revealing potential biomarkers and gene regulatory networks tailored to specific cell lines. Utilizing this data, we conducted a comparative analysis, pinpointing genes and pathways governing cellular traits. This informed our evaluation of therapeutic molecules like the curcumin analog EF-24. Our analysis of EF-24's effects on leukemia cell lines K-562 (ATCC® CCL-243™), HL-60 (ATCC® CCL-240™), THP-1 (ATCC® TIB-202™), and Kasumi-1 (ATCC® CRL-2724™) demonstrated increased cell death post-treatment. EF-24–activated pathways associated with cell survival and death regulation, notably impacting STAT1-induced cell death and enhancing innate immune system genes. This research offers a detailed understanding of EF-24's molecular actions in tumors, emphasizing the significance of STAT1 signaling in its antitumorigenic effects. The profound impact of EF-24 on inducing cell death across multiple leukemia cell lines underscores its potential as a therapeutic agent. By elucidating its specific molecular actions and highlighting the pivotal role of STAT1 signaling, this study not only deepens our comprehension of EF-24's mechanisms but also underscores its promising value in targeted antitumorigenic strategies.

Workflow for producing NGS data from ATCC cell lines

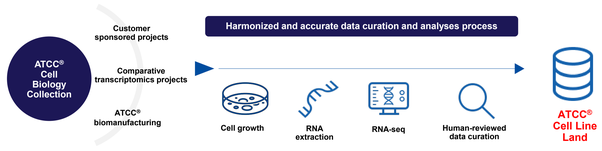

Figure 1: Schematic illustrating ATCC's workflow processes from cell culture to RNA extraction, sequencing, and data quality control in accordance with ISO 9001 standards.

RNAseq of ATCC's hematologic oncology cell models

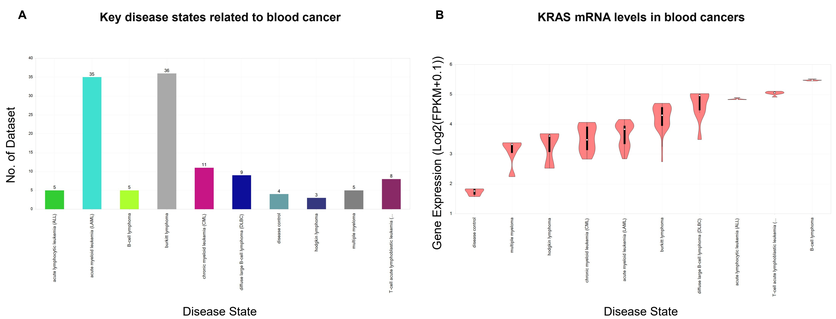

Figure 2: Whole-transcriptome profiling of hematological cancer cell models in the ATCC repository and the development of ATCC Cell Land, an authenticated data repository with analytical tools for bioinformatics analysis. (A) RNA sequencing of ATCC cell models representing various hematological disease states. The y-axis indicates the number of RNA sequencing data sets produced from biological replicates of the respective disease state. Numerous cell lines derived from each disease state were sequenced, each containing a minimum three biological replicates. (B) Box plots showing baseline mRNA levels of the KRAS gene in different types of blood cancer.

Landscape of gene expression in leukemia cell lines

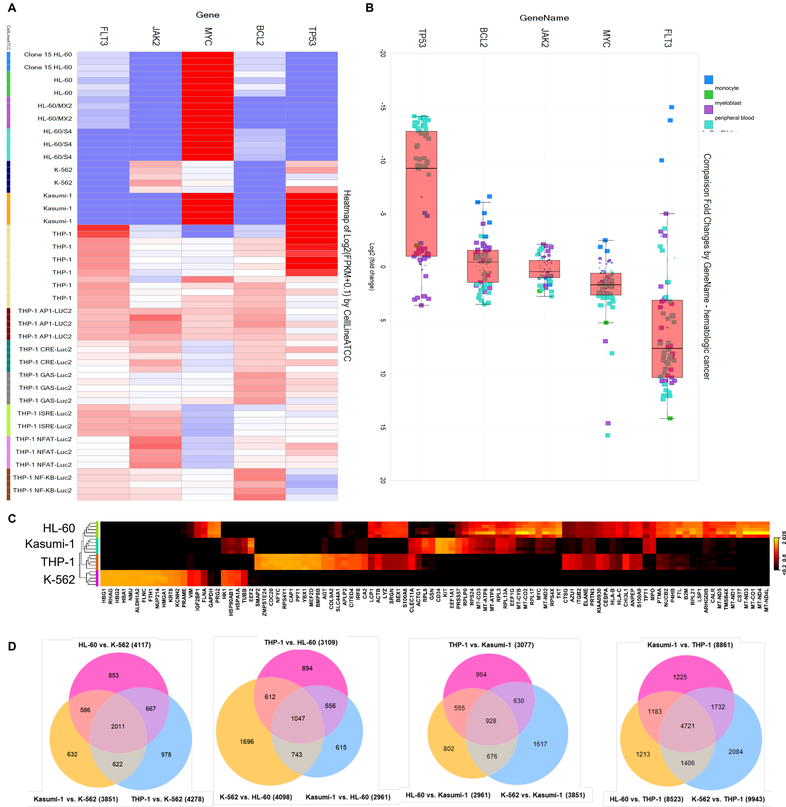

Figure 3: Comparative gene expression in leukemia disease states. (A) Expression profiles of hallmark genes in leukemia cell lines. (B) Box plots showing mRNA levels of the indicated genes in leukemia subtypes. (C) Heatmap depicting the transcriptomic landscape of leukemia cell lines. (D) Venn diagram illustrating the number of genes shared and uniquely expressed in leukemia cell lines. (E) Heatmap displaying genes with consistent expression patterns in THP-1, Kasumi-1, and HL-60 compared to K-562. Red indicates induction, while blue/black indicates reduction. The data reveal correlations (positive and or negative) among genes expressed in leukemia cell lines, highlighting their roles in disease pathogenesis.

Molecular modalities of EF-24 in leukemia cell lines

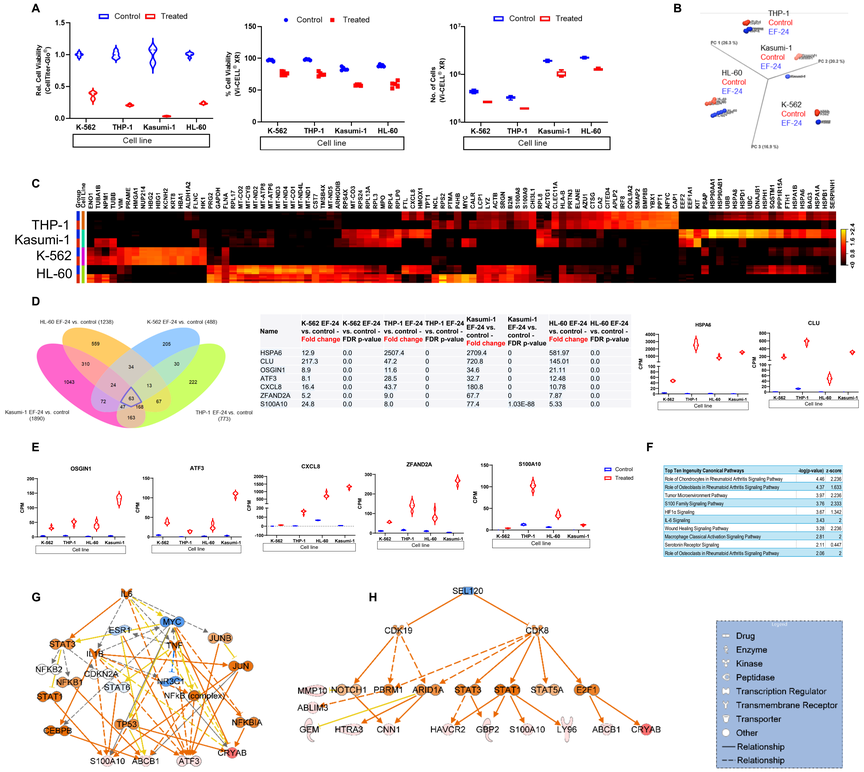

Figure 4: EF-24 exerts potent killing effects in leukemia cell lines. (A) Cell viability in leukemia cell lines is reduced after treatment with EF-24. Relative cell viability in control and EF-24 treatment groups as observed by the CellTiter-Glo (Promega Corporation) luminescent cell viability assay. (B) PCA plot illustrating sample clustering in leukemia cell lines and experimental groups. (C)) Heatmap of the top 100 DEGs in leukemia cell lines that shows the highest fold change in EF-24–treated cells as compared to untreated controls. There are 5 biological replicates of each condition/cell line (red = control; blue = treated). (D) Venn diagram depicts shared and unique genes that are differentially expressed in EF-24–treated cells as compared to untreated controls. Threshold absolute fold change ≥5, FDR p-value 0.01. (E) Pan differentially expressed genes in EF-24–treated cells as compared to untreated controls. (F) Violin plots display the relevant genes' quantitative enrichment in EF-24–treated cells as compared to untreated controls. (G) IPA shows shared top canonical pathways activated in myeloid leukemia cell lines were treated with EF-24. (H) IL-6 is an upstream regulator of genes induced in the EF-24 cell lines. (I) Regulatory network of genes induced in the EF-24 treated cell lines.

Summary

- ATCC Cell Line Land (ACL) provides comprehensive omics data for ATCC's verified and authenticated cell lines.
- It offers reference data for use as controls in experimental settings.
- Utilizing ACL data enhances reliability, accuracy, and reproducibility of research.
- ACL data enables gene expression analysis across multiple cell lines.
- It facilitates exploratory research and cell line screening against drugs.
- These data can be used as controls for publicly available data from ATCC® cell lines to ensure scientific rigor and accuracy.
- ACL is accessible through QIAGEN Digital Insights upon subscription. https://digitalinsights.qiagen.com/atcc-cell-line-land/

ATCC 10801 University Boulevard, Manassas, Virginia 20110-2209 **Phone:** 800.638.6597 **Email:** sales@atcc.org **Web:** www.atcc.org