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Blood Transcriptome Biomarkers

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Opinion

Blood transcriptomic profiling studies measure the abundance of blood transcripts on a genome-wide scale. Changes in blood transcript abundance reflect pathogenic processes and inform about the status of the immune system. Cancer biomarkers are biological molecules produced by the body or tumor in a person with cancer. Consequently, by analyzing the entire collection of RNA sequences in a cell (the transcriptome) researchers can determine when and where each gene is turned on or off in the cells and tissues of an organism. What can a transcriptome tell us? An RNA sequence mirrors the sequence of the DNA from which it was transcribed. Transcriptomics is the study of how our genes are regulated and expressed in different biological settings. Technical advances now enable quantitative assessment of all expressed genes.

A transcriptome is the full range of messenger RNA, or mRNA, molecules expressed by an organism. The term transcriptome can also be used to describe the array of mRNA transcripts produced in a particular cell or tissue type. Blood contains RNA within nucleated and enucleated cells as well as cell-free RNA, circulating in membrane vesicles. The RNA content is relatively low and varies between 1 to 5 bloods. This means that blood contains about 10 × more DNA than RNA. Because of this imbalance, a DNase digest is usually recommended during RNA isolation from human blood. Transcriptomics technologies are the techniques used to study an organism's transcriptome, the sum of all of its RNA transcripts.

The information content of an organism is recorded in the DNA of its genome and expressed through transcription. An exon is the portion of a gene that codes for amino acids. In the cells of plants and animals, most gene sequences are broken up by one or more DNA sequences called introns. RNA sequencing is the use of high throughput next generation sequencing technology to survey, characterize, and quantify the transcriptome of a genome. RNA sequencing has been used to analyze the pathogenesis of several malignancies such melanoma, lung cancer, and colorectal cancer. This step involves creating an index to evaluate the sequences for all possible unique sequences of length k (k-mer) in the transcriptome, which includes all known transcripts/ splice isoforms for all genes.

The index helps creates a signature for each transcript in our reference transcriptome. Transcriptome profiling is typically performed using hybridization or sequencing-based methodologies. Hybridization-based methods involve binding of fluorescently labelled fragments to complementary probe sequences either in solution or on a solid surface. DNA is usually extracted from one of two primary sources: cheek cells or white blood cells. Cheek cell samples carry an increased risk of contamination by viruses, bacteria or environmental elements.

Blood is, therefore, the preferred source of DNA samples. To increase RNA yields in (previously RNA-robust) tissue samples, avoid excessive homogenization or heat. Homogenizing in bursts of 30 seconds with 30-second rest intervals can improve RNA recovery. Also, eluting with more water releases more RNA from the membrane when using silica spin filters. Two biological techniques are used to study the transcriptome, namely DNA microarray, a hybridization-based technique and RNA-seq, a sequence-based approach. RNA-seq is the preferred method and has been the dominant transcriptomics technique since the 2010.

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