

Transcriptomics as a tool in studying cellular phenotype.

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Description

Transcriptomics is the study of RNA transcripts produced by a genotype at a given time, which provides a link between the genome, proteome, and cellular phenotype. It is a comprehensive approach that has evolved in recent years alongside genomics, proteomics, and metabolomics. Transcriptomics has been used in food microbiology to better understand microbial behaviour under various environmental conditions. It's also fascinating to learn about the overall response of a food's microbial community. Metatranscriptomics can be used to examine the collective transcriptome of the microbial community present in a food. With the advent of high-throughput sequencing technologies, transcriptomics has become an indispensable tool for unraveling the complex interplay between genes, their products, and their dynamic regulation in various biological processes and diseases.

Applications

The directions and potential applications of transcriptomics, including its integration with other omics approaches, its role in precision medicine, and its impact on drug discovery and development. We also highlight the ethical considerations associated with transcriptomics, such as data privacy, informed consent, and equitable access to technology. It gives an overview of the genetic content and the cell's regulatory system. The genome content of wild-type cells is the same whether they are bacterial cells or cell lines. Different genes are expressed in response to different applications and conditions, resulting in different patterns of gene expression in different organisms. There are cellular regulatory mechanisms that allow gene expression to be turned on and off. A great deal of research has been done on eukaryotes because they contain both introns and exons, which provide a better spectrum of the total transcriptome through splicing. Prokaryotic transcriptome research is lagging due to the absence of the 3'-end poly(A) tail, which is considered a signature of mature mRNA in eukaryotes.

Transcriptomics tools can assess data related to the expression level of genes in a given genome, genome profiling, comparative expression levels between different experimental data sets, and the effect of different parameters on gene expression. Techniques commonly used for transcriptome analysis include Expressed Sequence Tag (EST)-based methods, SAGE, hybridization-based microarray, real-time PCR, NGS-based RNA-sequencing (RNA-seq) methods, RNA interference, and bioinformatics tools. RNA isolation,

purification, quantification, cDNA library construction, and high-throughput sequencing are all part of the methodology. The following factors should be considered when selecting transcriptomics tools: cost effectiveness, sensitivity, high throughput, and minimal starting RNA concentration.

The sequencing of ESTs provides information about the gene's expression level. Large-scale EST sequencing projects on model organisms such as *Arabidopsis*, *Drosophila*, and humans have been carried out. There are also EST databases that can be used to reference the expression profile. Although it is useful for studying gene abundance and discovering new genes, it is an expensive and time-consuming technique. The cDNA and EST microarrays are available, with cDNA sequences and ESTs attached to a microchip. The method is similar to that of DNA microarray, but the sample here is fluorescently labelled cDNA molecules. It aids in the study of relative abundance between genes because it is a low-cost and quick technique. Another method, similar to EST sequencing, is SAGE.

It is superior to EST because only short "tags" of about 15 bases are sequenced. The generated short fragments are then joined together and sequenced. A pool of cDNA can be subjected to RNA-seq high-throughput NGS for quantification, discovery of novel ESTs, and RNA profiling. A DNA chip, gene chip, biochip, or microarray is a collection of DNA, cDNA, or oligonucleotide spots attached to a solid support such as glass or silicon chip to form an array for the purpose of expression profiling; monitoring the expression levels of thousands of genes at the same time; and is a hybridization-based method. Several advancements in chip fabrication, hybridization control, and computational analysis of generated data have been made over the years.

Techniques

Once the chip is manufactured, the high-density chip allows for relatively low-cost gene-expression profiling. The most significant limitation of this technology is that genome sequence information is required, as well as the higher background inherent in the hybridization technique. The quantitative real-time PCR (qRT-PCR) is a type of PCR that is commonly used for the accurate quantification of low-abundance mRNA or low-copy transcripts in transcriptomics studies. This technique's main advantages are its high sensitivity, better reproducibility, and wide dynamic quantification range. Because of its exponential amplification ability, it enables gene expression and regulation studies even in a single cell. Because of the availability of various types of

fluorescence monitoring systems attached to the PCR, it has grown in popularity for gene-expression studies. Some of the limitations of qRT-PCR include nonspecific amplification and the formation of primer-dimers. RNA-seq is a cutting-edge high-throughput transcriptomics technology. NGS tools are used in RNA-seq, also known as whole-transcriptome shotgun sequencing. The benefits of RNA-seq include the fact that it does not rely on the availability of the genome sequence, has no upper quantification limits, is highly reproducible, and has a wide dynamic detection range.

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