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Perspective

Sample Preparation and Lectin Microarray Hybridization of Glycomes

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DESCRIPTION

The term "glycomics" describes studies aimed at defining the complete range of glycans that a cell, tissue, or organism produces in specific time, location, and environmental conditions. This glycome's position on the cellular proteome is described by the term "glycoproteomics."

The genome, transcriptome, and proteome of a cell include data on the structure and complexity of the glycome. Therefore, no glycans in that cell can contain the glucose transferred by that glycosyltransferase at that specific time if a gene encoding a glycosyltransferase is not expressed (absent from the transcriptome). The complete glycome is presently difficult to predict due to the action of multiple glycosyltransferases and glycoside hydrolases competing for the same substrates in the biosynthetic pathway.

A technique for lectin-based glycan profiling is lectin microarray. It is a new platform for high-throughput analysis of glycans and glycoproteins that makes use of a panel of lectins immobilized on a clearly defined solid surface.

Through biochemical interaction analysis between the analyzed glycan and various lectins, lectin microarray can estimate an entire functionality of glycosylation of glycoproteins and cells. One of the most sensitive, efficient, and high-throughput profiling techniques, it allows for different lectin-glycan interaction assays to be performed simultaneously.

The most important advantage of lectin microarray is that it directly applies to glycoprotein glycans in both its pure and unprocessed forms (usually clinical samples like sera and tissue extracts). Lectin microarray has recognized as a special technique for determining the glycosylation properties of various glycoproteins.

Lectin microarrays include immobilizing lectins onto solid surface, glycan or glycoprotein sample labeling, sample binding to lectins and recognitions. There are different techniques for lectin immobilization, including carbine insertion, biotin-avidin bridge, attachment of amine functional group of lysine side chains of protein-backbone of lectins for immobilization to solid surface through epoxy-functionalized or N-hydroxysuccinimidyl (NHS)- determined esters, and 3D hydrogel surfaces.

The lectin sources can change from bacterial, parasitic, plant to synthetic, recombinant mutant libraries depending upon the purpose behind the lectin microarray assay. The glycoconjugates for binding can be from a different arrangement of analytes of biological samples, including glycoproteins, complete cell lysates, intact bacteria and even the entire mammalian cells. The glycans or glycoproteins are typically labeled with fluorescent tests. The predominant identification scheme in lectin array-based assays is the fluorescence detection.

The ratiometric approach is based on two-color labeling is also utilized in lectin microarray recognition for differential profiling of cellular glycomes. The lectin microarray in view of a evanescent-field of activated fluorescence detection enable to specific detection of fluorescently labeled glycans or glycoproteins without washing process and enables liquid stage observation in a equilibrium state.

As a high-throughput technology for glycan analysis, lectin microarray has turned into a promising methodology in glycomics and glycoproteomics. It can give rapid analysis of glycosylation profiles of glycoproteins and glycoform characterization. It is also utilized in methodology for quantitative analysis of lectin-glycoprotein interactions.

In comparative microarray analysis, structural differences can be detected as the changes in signal patterns of the lectin-binding intensities. Lectin microarray is a valid method for quality control of different glycoprotein products (such as, antibody drugs) and differential analysis of suitable clinical samples for the revelation of helpful glycan-related biomarkers. The bacterial cell-surface glycans and the mammalian cell-surface glycome both are analyzed using lectin microarray technology.

CONCLUSION

The complexity and apparently untameable nature of the glycome. Nature has developed lectins and glycan-binding proteins that perceive with intense selectivity. Synthetic small particles present new chances to interrogate and suppress the glycome. Small molecules that target glycans will naturally reveal

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new mechanisms for glycans control biological events. The representation of small molecules by their antagonistic effects toward development factor binding to glycosaminoglycans and their relating impacts toward mouse early stage immature microorganism separation. By utilizing this methodology, to enable discovery of Glycosaminoglycans (GAG)-binding particles that exhibit significant biological phenotypes.