Lectin Biosensors: An Alternative Tool for Pathogen Identification

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Abstract:

As there is a growing need for rapid, accurate and sensitive detection methods for pathogens use of biosensor is the best alternative tool for the same. Conventional methods for the microbial identification are sensitive but they usually require a long time for detection. Many of the new highly sensitive methods such as mass spectrometry and PCR based analysis require careful sample preparation and are also (rather) time consuming. Lectin are heterogenous class of proteins and glycoproteins that generally possess multiple subunits and have at least one oligosaccharide binding site per subunit. Several lectins are commercially available and their stability in standard buffers is better compared to monoclonal antibodies. Major advantage of using lectin is that the antibody can only be used to detect that antigen particularly while lectin as low affinity molecule may bind several different pathogens. The ability of lectin to react with microbial glycoconjugates means that it is possible to employ them as probes and sorbents for whole cells, and it makes them useful tools for identification or typing bacteria. Lectins are attractive reagents for the clinical diagnostic laboratory because of their diverse specificities, commercial availability, a wide range of molecular weight and their stability in standard buffers. The construction of lectin biosensors could be an advantageous method for detection of pathogenic bacteria.

Key words: lectin, biosensors, pathogens, antibodies

Introduction:

Detection of microorganisms is a key issue in fields like drinking water, quality assurance, food quality, biological terrorism threat control, applied medicine and fermentation technology (Agasti, 2010). For pathogen detection of very low concentration of microorganism is essential for an effective action, therefore there is a demand for efficient identification tool. According to recent study the best option is lectin biosensors.

Microbial surfaces bear many of the sugar residues capable of interacting with lectins. The ability of lectins to react with microbial glycoconjugates means that it is possible to employ them as probes and sorbents for whole cells, mutants and numerous cellular constituents and metabolites, and it makes them useful tools for identification or typing of bacteria. Since lectins and carbohydrates are both commonly present at the cell surface and because sugars posses tremendous coding capacity, they constitute excellent cell recognition markers that can be consequently exploited for therapeutic and diagnostic applications. In future genetic analysis of lectin producing gene may be useful to control pathogenic bacteria.

Sources of lectin:

Lectin can be obtained from Animal, Plant, and Microbial origin which could be either membrane bound or soluble intracellular. The biological role of lectin in the cell is not clear but it may be associated with sugar transport or carbohydrate storage in the cell. Some of them may be associated with attachment of symbiotic rhizobia to root nodules. As lectins have role in adhesion and agglutination, it is thought to be important in symbiotic as well as in pathogenic interaction among some pathogens and host.

Animal lectins:

Lectins have many different functions in animals like cell adhesion, regulation through glycoprotein synthesis, to control of certain blood protein level (Danguy, et al.1998). Lectins have also role in immune system by recognizing carbohydrates that are exclusive to pathogens (Karlsson, 1999). Carbohydrate recognition of lectins requires certain anomeric configuration and specific adjacent residues (Monzo et al., 2007). Lectins have also used to fractionate animal cells, like B cells T cells and lymphocytes. Some of the lectins can be obtained from the surface of mammalian liver cells that has specific role to recognize galactose residues.

Plant lectins:

For plants the proper function of plant lectin is not clear still. Best studied family of plant lectins is the leguminosae(e.g. Concanavalin-A, Soybean agglutinin and Lentil lectin). Most leguminous lectins are metalloproteins with tightly bound Ca⁺² and Mn⁺² which are essential for carbohydrate-binding activity. Some plant lectins, more properly classified

as toxins, however, are among the most poisonous proteins on our planet and can readily result in death not only to cells in cultures but also to animals. These toxic plant lectin subunits conjugated to specific antibodies and other targeting ligands are now being tested as treatments for cancer and other disorders of cellular proliferation (Slifkin and Doyle, 1990; Jordinson *et al.*, 1997; Takeya *et al.*, 1998).

Microbial lectins:

Microbial lectins play an important role in facilitating adhesion to the surface of microbial cells. Microbial lectins resemble plant lectins in carbohydrate specificity, relative thermostability, divalent cation requirements and other properties and may be used for some of the same purposes as other lectins that yield coaggregation responses with certain yeast cells (Slifkin and Doyle, 1990; Mirelman, 1986).

Principle of biosensor action and its components:

The principle of detection is the specific binding of the interest analyte or group of analytes (Figure-1) to the biorecognition element immobilized on a suitable support medium (Sharma *et al.*, 2003; Neethirajan *et al.*, 2005). This interaction is measured by transduction system then the signal is sent to read out display. Each interaction is result in specific change in one or more physico-chemical properties (pH change, electron transfer, mass change, heat transfer, uptake or release of gases or specific ions)are detected and measured by transducer (Neethirajan *et al.*, 2005). A biosensor generally consists of three main components as shown in Figure-1.

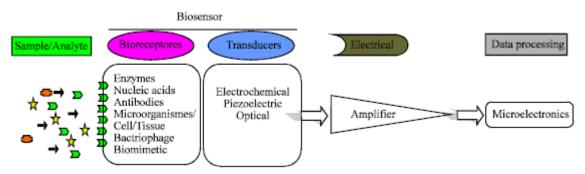


Figure 1: Principle of operation of a typical biosensor with its components

The biorecognition element, the transducer and the signal display or readout (Vo-Dinh and Cullum, 2000). The biological component of biosensor can be divided into two distinct groups, i.e., catalytic and non-catalytic. The catalytic group includes enzymes,

microorganisms and tissues, while the non-catalytic consists of antibodies, receptors, nucleic acids and some proteins such as lectins etc.

Various types of transducers are available for detection of analytes such as electrochemical (amperometric, potentiometric and conductometric), optical, colorimetric and acoustic etc. The biological materials especially enzymes, multi enzyme complex, tissues, microorganisms, organelles, cell receptors, antibodies, nucleic acids or whole cells (bacterial, fungal, animal or plant) are responsible for recognition of the analyte (Luong *et al.*, 1988).

Lectins for analytical applications:

As of today, lectins are normally chosen in bioseparation applications where glycoconjugates are involved. Figure-2 shows a typical method for the production of a lectin based optically detection approach.

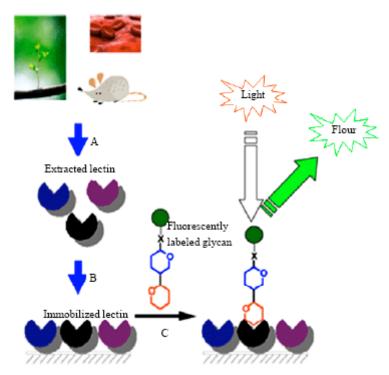


Figure-2: A typical method for the production of a lectin based optically detection approach including (A) lectin extraction, (B) immobilization on a solid surface and (C) biosensing of fluorescently labeled glycan

Different lectins are recommended to isolate glycoproteins and glycopeptides with distinct types of carbohydrate structures, viz. galectins are specific for N-acetyl-

lactosamine containing carbohydrates found in both N-linked and O-linked glycans (Kasai and Hirabayashi, 1996); Concanavalin A (Con-A) recognizes oligomannosyl motifs in N-linked-glycans (Ohyama *et al.*, 1985); peanut agglutinin is specific for O-linked-glycans (Neurohr *et al.*, 1980) and *Aleuria aurantia* lectin shows broad specificity for fucose-containing oligosaccharides (Kochibe and Furukawa, 1980; Monzo *et al.*, 2007), Which indicates potential use of Lectins for biosensing purposes and biosensor technology.

Medical samples analysis:

The most important applications of lectin-based biosensors are in the field of medicine and therapeutics. Many researches have already been done in this field and a lot of such sensors have been developed. Also, the large amount of published articles about the applications of lectin in the assessment process of clinical samples, laboratories and diagnostic instruments can prove the importance of this issue. Actually, the applicability of biosensing systems for precise determination of analytes of biomedical interest from diverse physiological clinical samples such as blood, urine, saliva, cell tissues, etc. is highly desirable to establish quick and reliable analytical tools avoiding time-and sample-consuming pretreatment methods (Shankaran *et al.*, 2007).

Conclusion and prospective:

The diagnostics market is expanding rapidly and entering a wide range of disciplines including agricultural and food industry, medicine and environment monitoring. It seems that biosensors could play an important role as a powerful analytical tool in the diagnostic sectors, particularly where rapid, low cost, high sensitive and specific measurements in field situations are required. There are different approaches to combine biology, chemistry and electronics in order to develop new biosensors with different applications in diagnostic field. Lectin proteins are one of the most attractive biorecognition elements for providing biosensors. It is anticipated that lectin-affinity-based separation and detection methods and instruments, especially biosensors, will maintain their importance in diagnostic researches. New techniques also continue to be developed in order to study lectins and to utilize their specificities in functional assays.

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