GlycoSeLect

Bio-Layer Interferometry with Recombinant Prokaryotic Lectins

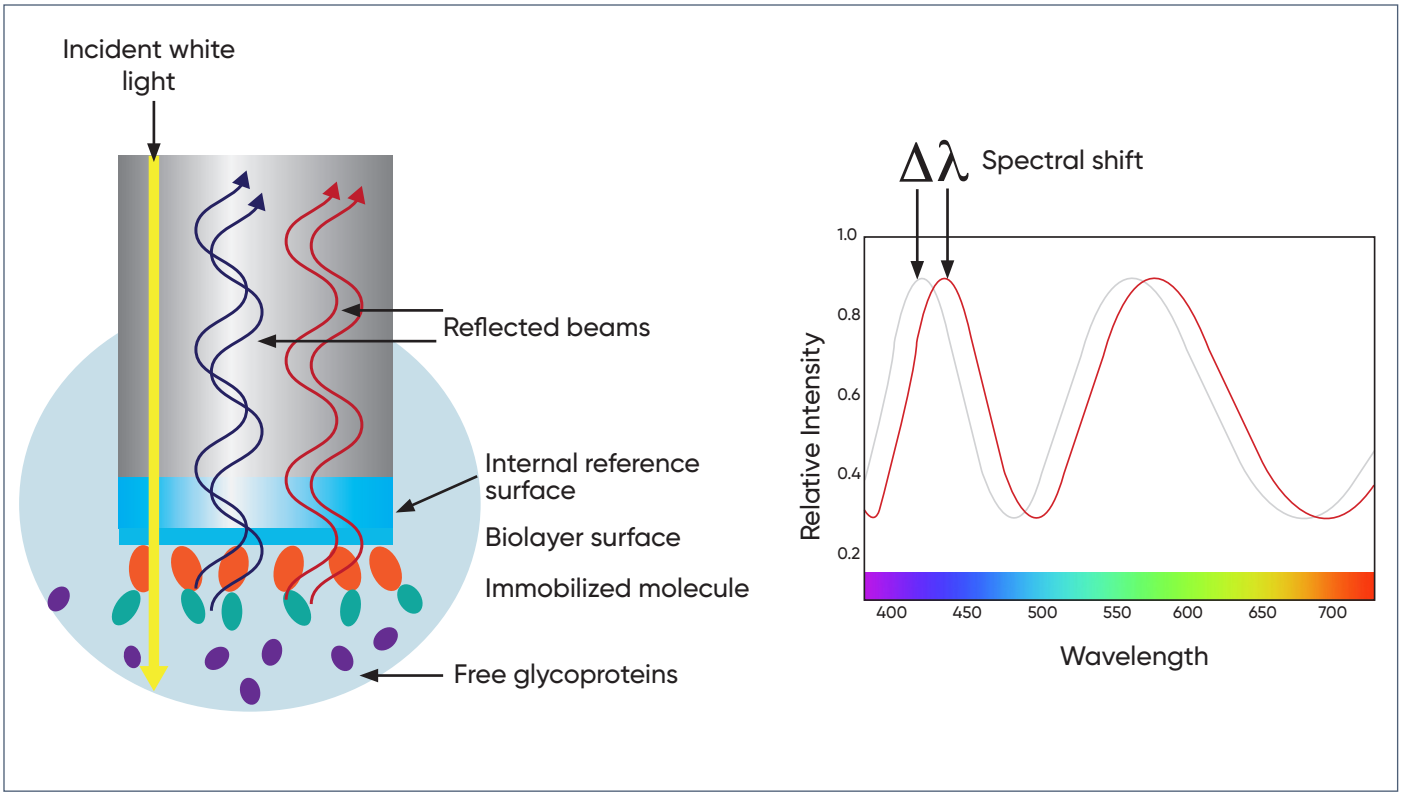
RPL biosensors enable rapid, high throughput glycoanalysis in real-time

Bio-Layer Interferometry (BLI) is widely used for studying biomolecular interactions such as antibody binding to an antigen, or small molecule drug binding to a protein target. It has also been adapted for glycoanalysis, making it a valuable tool for applications spanning basic research through to biotherapeutic manufacturing. Recombinant Prokaryotic Lectins (RPLs) have proven utility for BLI, where they offer several advantages over traditional, plant-based lectins.

What is Bio-Layer Interferometry?

Bio-Layer Interferometry is a label-free technology for monitoring the interaction between an immobilised target and a ligand free in solution. It uses an optical biosensor comprising two distinct surfaces - an internal reference surface and a biolayer surface – which differentially reflect white light to generate a specific interference spectrum. When a ligand binds to the biolayer surface, it increases the optical thickness, causing a shift in the interference spectrum that can be measured in real time. By comparing this readout to a reference (ligand-free) biosensor, researchers can investigate the binding affinities and kinetics of many different biomolecular interactions.

Bio-Layer Interferometry assay principle



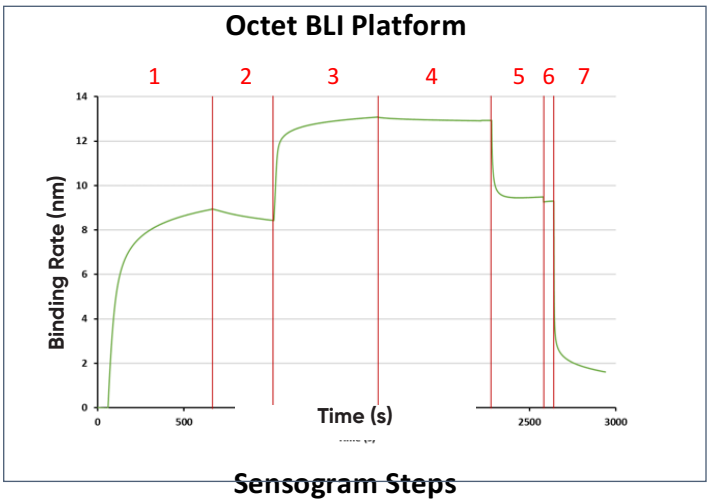
What are Recombinant Prokaryotic Lectins?

Lectins are a class of proteins that bind glycans, the chain-like structures of single sugar molecules (monosaccharides) produced by almost all living organisms for various structural, protective, and regulatory purposes. They have traditionally been purified from plants; however, plant-based lectins lack specificity, making their use for certain applications challenging. Recombinant prokaryotic lectins are instead expressed as soluble proteins by E. coli, giving them several important advantages over plant-based alternatives.

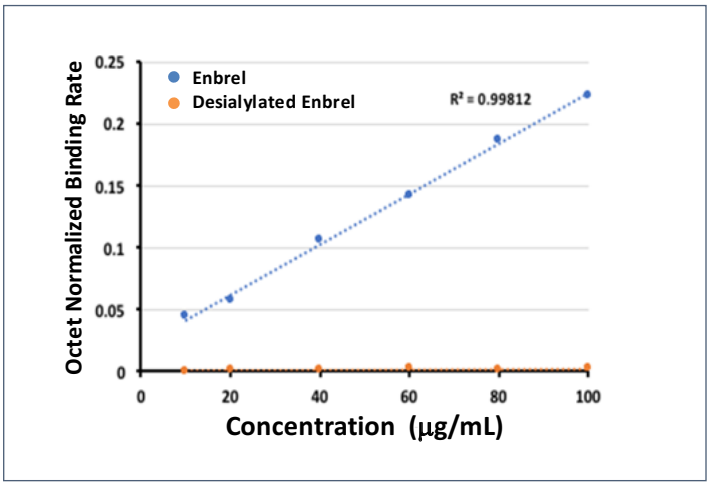
- Superior specificity
- Consistent batch-to-batch performance
- Simple, scalable production
- Open to engineering – can enhance glycan binding or enable the expression of protein tags

How are Recombinant Protein Lectins used for BLI?

RPLs can be immobilised on the surface of BLI biosensors and used for evaluating the glycosylation status of proteins of interest. Being able to perform glycoanalysis in this way improves on conventional HPLC and MS-based approaches by reducing experimental complexity and increasing throughput. This is especially beneficial for biopharmaceutical manufacturing, where glycosylation represents a Critical Quality Attribute (CQA) that must be closely monitored due to its potential to influence protein structure and function.



Typical RPL biosensor data:
1 - RPL immobilisation
2 - equilibration
3 - glycoprotein association
4 - dissociation
5 - glycoprotein elution
6 - equilibration
7 - biosensor regeneration



Glycoanalysis of Enbrel:
Sialic acid and galactose specific RPLs were immobilised on biosensors for glycoanalysis with the Sartorius Octet.

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