Glycosciences Products 2019

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Applied Glycomics

Sommaire

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Lectins

Description

Lectins are proteins or glycoproteins which possess the ability to bind specifically sugars. They have no enzyme activity and are not antibodies. Lectins are ubiquitous in nature, being found in all kinds of organisms (virus, microorganisms, plants, invertebrates and vertebrates). Lectins are usually oligomeric proteins and have many binding sites. The binding constant of the specific free sugar is generaly many orders of magnitude lower than the binding constant of a glycoconjugate (glycolipid, glycoprotein...) containing this sugar. Lectins agglutinate cells, some lectins are even blood type specific, but they are also able to recognise cells surface glycans allowing to distinguish between different cells species and states. Furthermore some lectins stimulate lymphocyte and induce mitosis. The lectins have been used for :

- **Studies of glycobiological interactions with glycans or glycans mimics**
- **Detection, isolation, and structural studies of glycoproteins**
- **Study the dynamics of the cell surface glycoconju gates**
- **Cell identification and to separate subpopulation of cells and subcellular organelles**
- **Study endocytosis, neoplastic transformation**
- **Mitogenic stimulation of lymphocytes**
- **Glyco-biomarkers dicovery and new diagnostics assays design**

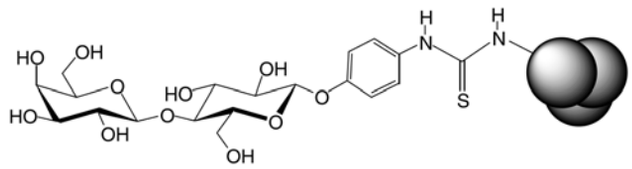
Reference	Short Name	Common name	Glycans structures specificity
		Natural Lectins	
L1222	ABA	*Agaricus Bisporus*	Gal(β-1,3) GalNAc
L1221	AIA / Jacalin	*Artocarpus intergrifolia*	Gala1-6 or Galb1-3GalNAc (T-antigen)>> lactose, more specific for T-antigen than PNA
L1367	AML	*Astragalus membranaceus*	Galb
L1205	ASA	*Allium Sativum agglutinin*	α(1,3)-linked mannosyl units
L1889	BanLec	*Musa Acuminata*	αMan
L1254	CJA	*Crotalaria juncea*	Gal (Lac>GalNAc)
L1366	cMOL	*Moringa oleifera*	Complex glycans, inhibited by asialofetuin
L1201	Con A	*Canavalia ensiformis*	Man > Glc ; branched mannoses a
L1206	GNL / GNA	*Galanthus nivalis*	Terminal mannoses. Mana1-3Man ; a2-macroglobulin ; bind mannopentaose
L1202	LcH	*Lens culinaris*	Mana/Glca > GlcNAca, enhanced by Fuca1-6 on the core GlcNAc-Asn N-glycopeptides
L1252	NPA	*Narcissus pseudonarcissus Daffodil*	External or internal a or b mannose
L1240	PHA E	*Phaseolus vulgaris*	Galb1-4GlcNAcb1-2Man, the bisecting GlcNAcb1-4Man is essential.
L1239	PHA L	*Phaseolus vulgaris*	Galb1-4GlcNAcb1-6Man of branched structures of N-glycans, Galb1-4GlcNAcb1-2Man.
L1585	PHA M	*Phaseolus vulgaris*	mix of PHA-E and PHA-L specificities.
L1586	PHA P	*Phaseolus vulgaris*	mix of PHA-E and PHA-L specificities.
L1223	PNA	*Arachis hypogaea*	Lactose, T- antigen
L1203	PSA, PEA	*Pisum sativum*	Man > Glc ; enhanced by Fuca1-6 on the core GlcNAc-Asn N-glycopeptides, IgM1A mouse
L1216	SBA	*Glycine max*	Preference for a over b-glycodidic linkage.
L1237	SNA	*Sambucus nigra*	Neu5Aca2-6Gal/GalNAc
L1476	TJA-II	*Trichosanthes japonica Agglutinin II*	Fuca1-2Galb-, GalNAcb-, Galb1-3/4-GlcNAc-, Galb1-6Gal-
L1261	TXLC-I	*Tulipa gesneriana agglutinin*	GalNAc, Gal
L1253	VEA	*Vicia ervilia*	Man>trehalose>Glc
L1230	WGA	*Triticum vulgare*	GlcNAc; GlcNAcb1-4 oligomers , core of Asn linked oligasacchide; Neu5Ac
		Recombinant Lectins	
L1255	BC2L-A	*Burkholderia cenocepacia lectin A*	Manα1-2, Manα1-3, Manα1-6, dimanoside,
L1257	PA-IL	*Pseudomonas aeruginosa lectin A*	Galα, Globoside
L1256	BC2L-C	*Burkholderia cenocepacia lectin C (N terminal domain)*	Fuc, oligo H type I, Lewis B, Lewis Y
L1258	RSL	*Ralstonia solanacearum*	Fuc
L1259	PAII-L	*Pseudomonas aeruginosa lectin B (Lec B)*	Fuc >> Man, Lewis A

Neoglycoproteins

Description

Neoglycoproteins are **glycosylated bovine serum albumin** (BSA) molecules obtained after the conjugation of a phenylisothiocyanate glycosides with the ε-amino groups of lysine residues of BSA. The synthesis of each neoglycoprotein is conducted under a standardized procedure allowing an excellent batch to batch reliability. Each neoglycoprotein is submitted to a complete quality control ensuring a total conformity with the specifications : purity, carbohydrates/protein ratio, labeling and **functionnality assessed by interactions with lectins**.

Mono and di-saccharide neoglycoproteins are **produced routinely and always available (from 0.5 mg to 50 mg)** in unlabeled or fluoresceinylated forms. **Biotinylated or other conjugates as well as more complex neoglycoproteins** are available upon request.



Intended use

Neoglycoproteins are known as “amplifiers” of carbohydrates-proteins interactions. The use of neoglycoproteins as tools to decipher glycoconjugates, carbohydrates binding proteins and more generally proteins-carbohydrates interactions were described in many studies (see bibliography). Neoglycoproteins are used in number of methods including histochemistry, ELISA assays, blotting assays, affinity chromatography, cytochemistry by flow cytometry, confocal or electron microscopy.

Neoglycoproteins can be use for research purposes to:

- **Identify lectins or lectin-like proteins.**
- **Purify lectins or other carbohydrate-binding proteins.**
- **Design new diagnostic tools**.
- **Discover biomarkers.**
- **Target drugs**.
- **Trigger immune response** against carbohydrates moieties.

Benefits

- The **affinity** of the neoglycoproteins is 10_2-10_4 higher than that of the corresponding free sugars.
- The neoglycoproteins are very reliable and **stable products** that can be labeled with great flexibility.
- The **high solubility** in aqueous solutions makes neoglycoproteins very powerfull reagents for glycosciences studies.

Description	Reference
βChitobiose-BSA	NeoCT
βchitobiose-BSA-F*	NeoCTF
αLFuc-BSA	NeoF
αLFuc-BSA-F	NeoFF
αDGal-BSA	NeoGa
αDGal-BSA-F	NeoGaF
βDGal6P-BSA	NeoGaP
βDGal6P-BSA-F	NeoGaPF
αDGalNAc-BSA	NeoGaN
αDGalNAc-BSA-F	NeoGaNF
αDGlc-BSA	NeoaG
αDGlc-BSA-F	NeoaGF
βGlc-BSA	NeobG
βGlc-BSA-F	NeobGF
βDGlcNAc-BSA	NeoGN
βDGlcNAc-BSA-F	NeoGNF
βDLac-BSA	NeoL
βDLac-BSA-F	NeoLF
αDMan-BSA	NeoM
αDMan-BSA-F	NeoMF
αDMan6P-BSA	NeoMP
αDMan6P-BSA-F	NeoMPF
αLRhamnose-BSA	NeoR
αLRhamnose-BSA-F	NeoRF
BSA-F	NeoBF
Glucitol-Bsa-F	NeoGolF

* : F = Fluoresceinylated.

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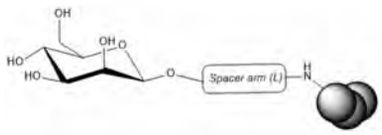
Neoglycoclusters

Description

Neoglycoproteins are **glycosylated bovine serum albumin** (BSA) molecules. Our first range of simple neoglycoproteins was achieved by the conjugation of phenylisothiocyanate glycosides with the ε-amino groups of lysine residues of BSA.

In order to improve accessibility and avidity of a carbohydrate-binding proteins, a new version of neoglycoproteins containing spacer arm (*i.e.* an alkyl spacer or a polyethylene glycol (PEG) chain), were developped and proposed either with monosaccharides / disaccharides or with glycoclusters

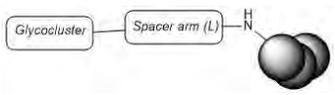
- Monosaccharide spacer neoglycoproteins:



Description	Reference
αDMan-BSA	NeoM_A_O1
αDMan-BSA	NeoM_P_O1
αDMan-BSA	NeoM_P_2O1
αLFuc-BSA	NeoFuc_A_O1
αDGal-BSA	NeoGa_A_O1

A = linker alkyle ; P = linker PEG
O1 = 1 monosaccharide/arm ; 2O1 = 1 disaccharide/arm

- **Neoglycoclusters**, achieved by introduction of a carbohydrate cluster containing 3 to 9 carbohydrates units:



Description	Reference
αDMan-BSA	NeoMClus_O3
αDMan-BSA	NeoMClus_O9
αDMan-BSA	NeoMClus_2O3
αDMan-BSA	NeoMClus_2O9

O3 = 3 monosaccharides/cluster; 2O3 = 3 disaccharides/cluster
O9 = 9 monosaccharides/cluster ; 2O9 = 9 disaccharides/cluster

The synthesis of each neoglycoprotein and neoglycocluster is conducted under a standardized procedure allowing an excellent batch to batch reliability. Each neoglycoprotein and neoglycocluster is submitted to a complete quality control ensuring a total conformity with the specifications: purity, carbohydrates/protein ratio, labeling and **functionnality assessed by interactions with lectins through GLYcoPROFILE method**.

Monosaccharide spacer neoglycoproteins and neoglycocluster are **produced routinely and always available (from 0.5 mg to 1 mg)** in unlabeled forms *(labeled products available on request).*

Intended use

Neoglycoproteins are known as “amplifiers” of carbohydrates-proteins interactions. The use of neoglycoproteins as tools to decipher glycoconjugates, carbohydrates binding proteins and more generally proteins-carbohydrates interactions were described in many studies (see bibliography).

Neoglycoproteins can be use for research purposes to:

- **Identify lectins or lectin-like proteins.**
- **Purify lectins or other carbohydrate-binding proteins.**
- **Design new diagnostic tools**.
- **Discover biomarkers.**
- **Target drugs**.
- **Trigger immune response** against carbohydrates moieties.

Benefits

- The **affinity of neoglycocluster** is 10^2 to 10^3 higher than usual neoglycoprotein.
- Neoglycoproteins ans neoglycoclusters are very reliable and stable compound.
- The high solubility in aqueous solutions makes neoglycoproteins and neoglycocluster very powerfull reagents for glycosciences studies.

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- Duverger *et al.* (1999). Interaction between lectins and neoglycoproteins containing new sialylated glycosynthons. *Glycoconjugate J.*, **16**, 793-800.
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- Monsigny *et al.* (2007). Carbohydrate-mediated Interactions. 3.23. Neoglycoproteins. Comprehensive Glycoscience. From Chemistry to Systems Biology. Amsterdam, Elsevier. **3**, 477-521.

LEctPROFILE plate

Description

The **LEctPROFILE plate** is a lectin array (1,2) proposed by GLYcoDiag to highlight specific types of structures and/or to indicate the potential modifications of glycans with respect to reference structures. The relevant choice of a range of lectins makes it possible to validate the structure of glycans in a short time and with very simple basic equipment.

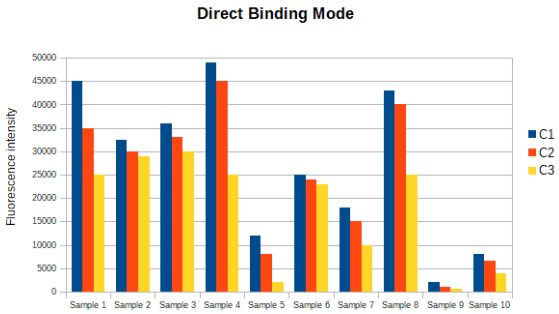
Each lectin are immobilized on the bottom of microtiter plates (96-well format), intended for absorbance or fluorescence interaction measurements. Up to 28 different lectins (see the list below) are proposed in a minimum format of 2 strips of 8 wells, in order to compose one or more microplates adapted to the desired analysis.

code	Short name	Common name	code	Short name	Common name
LP1251	GSL II	*Griffonia simplicifolia*	LP1523	PNA	*Arachis hypogaea*
LP2271	PWM	*Phytolacca americana*	LP1633	LTA, LTL	*Lotus tetragonolobus*
LP3301	WGA	*Triticum vulgare*	LP1734	AAL	*Aleuria aurantia*
LP4811	ACA, ACL	*Amaranthus caudatus*	LP1844	UEA I	*Ulex europeus*
LP5911	BPA	*Bauhinia purpurea*	LP1936	MAA	*Maackia amurensis*
LP1011	DBA	*Dolichos biflorus*	LP2037	SNA	*Sambucus nigra*
LP7131	HPA	*Helix pomatia*	LP2139	PHA L	*Phaseolus vulgaris*
LP8461	PTL-I	*Psophocarpus tetragonolobus*	LP2240	PHA E	*Phaseolus vulgaris*
LP9161	SBA	*Glycine max*	LP2349	CorM	*Coregonus lavaretus marenae*
LP1019	WFA	*Wisteria floribunda*	LP2411	Con A	*Canavalia ensiformis*
LP1121	AIA / Jacalin	*Artocarpus intergrifolia*	LP2521	LcH	*Lens culinaris*
LP1241	GSL-Ib4	*Griffonia simplicifolia isoB4*	LP2631	PSA, PEA	*Pisum sativum*
LP1342	MOA	*Marasmius oreades agglutinin*	LP2761	GNL / GNA	*Galanthus nivalis*
LP1422	ABA	*Agaricus bisporus*	LP2871	HHL, HHA	*Hippeastrum hybrid*

Table 1. Lists of lectin available for the LEctPROFILE plate.

Applications

The evaluation of compounds interactions with lectins is achieved by the **direct binding mode** that evaluate potential interaction of compounds ranging from pure molecule to complex mixtures (glycocojugate(s), complex carbohydrates or glycomimetics). Previous labeling of target molecule(s) by biotinylation or by fluoresceinylation is required for readout.



Name	Content	Analysis mode	Stability
LEctPROFILE plate	2 x 8 well strip per lectin used for fluorescence or absorbance detection	*Direct Binding:* until 10 samples analysed in triplicate at 3 concentrations	Each LEctPROFILE plate are stable for minimum 6 months at -20 °C

Table 2. Specifications of LEctPROFILE plates.

Bibliography

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2. Hirabayashi, J., Yamada, M., Kuno, A., Tateno, H., *Lectin microarrays: concept, principle ans applications*, Chem. Soc. Rev., **2013**, *42*, 4443-4458.

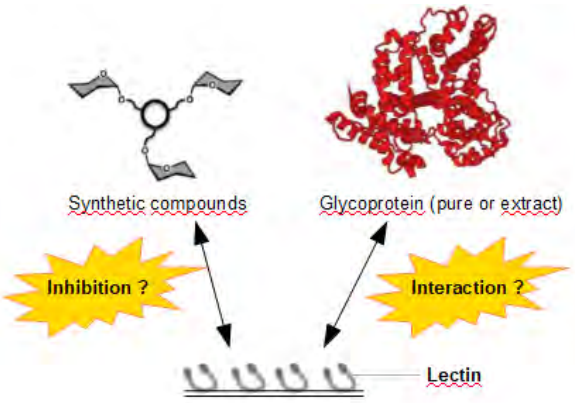
LEctPROFILE kit

Description

The **LEctPROFILE kit** allows **efficient** evaluation of crude or purified glycoconjugates interactions (*i.e.* synthetic molecules or glycoconjugates) with lectins by a simple measurement of absorbance or fluorescence. The LectPROFILE kit enables a **fast measurement** (below 3 h) and are **easily accessible to all**.

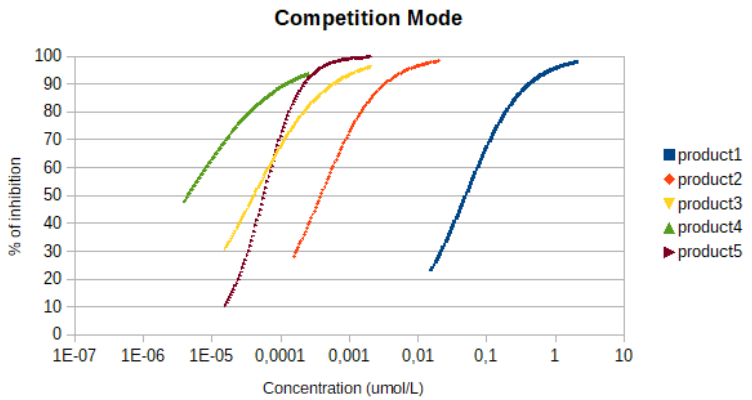
Each kit are composed of a 96-well plate immobilised with the target lectin, the corresponding tracor and the revealing solution.

All our lectins are controlled under a standardized procedure assessed by interactions with specific neoglycoproteins or glycoproteins through GLYcoPROFILE method.



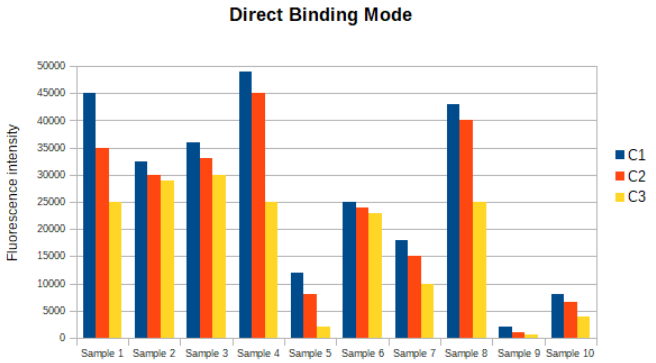
Applications

The evaluation of compounds interactions with lectins is possible by two different modes:



- Competition Mode : Competitive interaction between a sample and a specific labelled tracer known to have good affinity for the lectin without preliminary labeling of sample. This mode is typically used for the evaluation of the IC50-value (*i.e.* concentration corresponding to 50% of lectin inhibition), for the screening of potential candidate, for avidity comparison or for batch to batch monitoring.

- Direct Binding are used to evaluate potential interaction of compounds ranging from pure molecule to complex mixtures (glycocojugate(s), complex carbohydrates or glycomimetics). Previous labeling of target molecule(s) by biotinylation or by fluoresceinylation is required for readout.



Name	Specificity	Kit Content	Analysis mode	Stability
FimH[1] LEctPROFILE kit	High mannosylated structure glycan(s)	1 x 96 microplate well for fluorescence detection; Assay reagents: FimH tracor & Streptavidine-DTAF solutions	*Competition Mode: until 5-8 samples analysed in triplicate* *Direct Binding:* until 10 samples analysed in triplicate at 3 concentrations	6 months at -20 °C
LEctPROFILE kit	-[a]	1 x 96 microplate well for fluorescence or absorbance detection; Assay reagents: lectin tracor & revealing solution	*Competition Mode: until 5-8 samples analysed in triplicate* *Direct Binding:* until 10 samples analysed in triplicate at 3 concentrations	-[b]

Table 1. *Specifications of LEctPROFILE kit. a. For specificity of LEctPROFILE kit, see our lectins specificities table. b. Each LEctPROFILEs kits are stable for minimum 6 months at -20 °C.*

Bibliography

1. Hartmann, M.; Lindhorst T. K.; *Eur.J.Org.Chem.*, **2011**, 3583-3609.

Affinity gel chromatography

Description

Affinity gel chromatography[1] which aim to purify molecule by non-covalent and reversible binding is in the most cases the method of choice to obtain desired lectins or conversely oligo-polysaccharides or glycoconjugates with high yield and purity. These reagents are provided either as bulk or prepacked into 1 to 5 mL columns.

Two kind of affinity gel are proposed by GLYcoDiag with different specificities:
Carbohydrate affinity gel (CarbPROFILE gel) or Lectin affinity gel (LEctPROFILE gel).

Carbohydrate affinity gel (CarbPROFILE gel)[2]

CarbPROFILE gels are monosaccharides-Sepharose affinity matrices used for purification of specific carbohydrate-binding proteins. The carbohydrates are attached through their non reducing hydroxyl group after pre-activation of sepharose matrix by divinylsulfone (DVS) *(see scheme 1 below)*. The binding of lectins and carbohydrates binding proteins to carbohydrate affinity gel is non-covalent and reversible with high capacity. Lectins and carbohydrates binding proteins are both usually stable compounds which can be recovered by competitive elution (*i.e.* 0.2 to 0.5 M of monosaccharide) or by modulations of pH and/or ionic strength in high yield and purity.

Scheme 1. *CarbPROFILE gel matrix.*

Reference	Name	Specificity	Capacity *(mg of protein/mL of gel)*	Unit size[a] *(mL)*
ManGel	Mannose-CarbPROFILE gel	Mannose binding protein	> 30 (based on ConA lectin)	5, 10 or 25
GalGel	Galactose-CarbPROFILE gel	Galactose binding protein	> 15 (based on AIA lectin)	5, 10 or 25
GlcNAcGel	N-Acetylglucosamine-CarbPROFILE gel	N-Acetylglucosamine binding protein	> 15 (based on WGA lectin)	5, 10 or 25

Table 1. Specifications of CarbPROFILE gel. a. *available in a pre-packed column or in suspension.*

Applications

- Purification of large range of glycoproteins which recognize specific carbohydrates moeities[3].

Lectin affinity gel (LEctPROFILE gel)

LEctPROFILE gels are affinity gel chromatography where lectins are immobilized on a Sepharose 4B fast flow matrix. Glycoconjugates can be recovered by competition with the specific inhibitory monosaccharide of the lectin. LEctPROFILE gels are used for the purification of glycoconjugates with specific N-glycan residues. LectPROFILE gel are personnalized reagents produced on your request *(see our list of available lectins)*.

Scheme 2. LEctPROFILE gel matrix

Name	Specificity	Binding capacity *(mg of glycoprotein/mL of gel)*	Unit size[a] *(mL)*
LEctPROFILE gel	-[b]	≥ 1	1, 2 or 5

Table 2. Specifications of LEctPROFILE gel. *a. available in a pre-packed column or in suspension; b. For specificity of LEctPROFILE gel, see our lectins specificities table (available on our website, www.glycodiag.com).*

Applications

- Purification of polysaccharides.[4]
- Purification of complex glycoproteins having specific glycans structures.[5]

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1. Pohleven, J., Štrukelj, B., Kos, J. *Affinity Chromatography of Lectins, Affinity Chromatography,* 2012, IntechOpen.
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5. Sueyoshi, S., Tsuji, T., Osawa, T., *Biol. Chem. Hoppe-Seyler,* **1985**, *366*, 213-221.