

SiaFind[™] Pan-Specific Lectenz[®] 2.0 Kit contents

Catalog #	Description	Size	M. W.	Storage
SK0502	SiaFind [™] Pan-Specific Lectenz [®] 2.0 (SP0503-1MG)	1 mg, lyophilized	57,000	-20°C, up to 12 months
	5X SiaFind™ Binding Buffer 2 (BA0102)	100 mL		4 to 25°C
SK0502B	SiaFind™ Pan-Specific Lectenz® 2.0, Biotinylated (SP0503B-1MG)	1 mg, lyophilized	58,000	-20°C, up to 12 months
	5X SiaFind™ Binding Buffer 2 (BA0102)	100 mL		4 to 25°C

This product is for research use only and not for resale or for any use in the manufacture of a therapeutic or for any diagnostic purpose.

Product Description

Lectenz[®] are a novel class of <u>lect</u>in-like, <u>enz</u>yme-derived glycan-targeting affinity reagents engineered by computationally-guided directed evolution. The reagents are highly purified recombinant proteins, each designed to bind a specific glycan structure, and have advantages over naturally occurring lectins in rapid detection and enrichment of glycoconjugates.

SiaFind[™] Pan-Specific Lectenz[®] 2.0 Kits (Cat #SK0502 and SK0502B) contain a high-performance sialic acid affinity reagent (SP0503) engineered from the original SiaFind[™] Pan-Specific Lectenz[®] (SP0502) for the robust detection, separation, or enrichment of sialoglycans terminating in Siaα2,3Gal, Siaα2,6Gal, and Siaα2,8Sia commonly found in glycoconjugates (glycoproteins, glycolipids, and oligo- or polysaccharides). It has high affinity and specificity towards sialoglycans in a linkage independent manner (pan-specific). Each kit also includes a 5X binding buffer to ensure maximum reagent specificity and ease of use.

SiaFind™ Pan-Specific Lectenz® 2.0 has a molecular mass of about 57 kD and works as a monomer without bivalent metal ions. It is 8xHis-tagged at its *N*-terminus, and an anti-polyhistidine antibody, or in the case of the biotinylated version, a streptavidin conjugate can be used for detection. The 8xHis tag may be removed by FasTEV™ (Cat #GE0501), a TEV protease with enhanced stability and catalytic activity.

Form and Storage

The **SiaFindTM Pan-Specific Lectenz® 2.0** are supplied lyophilized in a storage buffer (50 mM EPPS, 200 mM NaCl, pH 7.5) and should be reconstituted in 100 μ L molecular grade water to yield a 10 mg/mL solution. Concentration is determined by spectrophotometry using E^{1%} 10.4. Once reconstituted, store at 4°C for up to 5 days or -20°C for up to 6 months. Aliquoting is recommended to avoid repeated freeze-thaw cycles.

All 5X buffers should be diluted to 1X with ultrapure water. For instance, to make 250 mL, add 50 mL of any 5X buffer to 200 mL water and mix by inversion. All buffers may be stored at 4 to 25°C.

Western Blotting Guide

Use 0.1 - 1.0 μ g fetuin and/or sialyllactose-BSA as positive control.

Prepare 1X SiaFind[™] Binding Buffer 2 (SBB2, 25 mM EPPS, 100 mM NaCl, pH 7.5) from the 5X binding buffer (Cat #BA0102). Prepare SBB2 plus 0.1% Tween-20 (SBB2T) for membrane washing.

Prepare SBB2T with 5% globulin-free BSA for blocking. Incubate the membrane at room temperature for 1 h with agitation.

Prepare **SiaFind**TM Lectenz[®] in SBB2T with 0.5% globulin free BSA: 10 μ g/mL of the native reagent (Cat #SP0503) or 2.5 μ g/mL of the biotinylated reagent (Cat #SP0503B). Incubate at room temperature for 1 h with agitation. Rinse membrane 3 X 5 min with SBB2T.

Incubate with a 2° probe diluted in SBB2T with 0.5% globulin-free BSA, e.g., a 1:10,000 dilution of an anti-polyhistidine tag antibody-HRP conjugate for the native reagent (Cat #SP0503) or 1 μ g/mL of a streptavidin-HRP solution for the biotinylated reagent (Cat #SP0503B). Rinse membrane 3 X 5 min with SBB2T.

Rinse membrane 3 X 5 min with SBB2 before applying HRP chemiluminescent substrate for detection.

Note: SiaFind™ Pan-Specific Lectenz® 2.0 will work in common Western Blotting buffers, such as TBS, with slight reduction in binding signal.

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