

SiaFind™ α 2,3-Specific Lectenz® Kit contents

| Catalog # | Description | Size | M. W. | Storage |
|-----------|---|-------------------|--------|------------------------|
| SK2301 | SiaFind™ α 2,3-Specific Lectenz® (SP2302-1MG) | 1 mg, lyophilized | 77,000 | -20°C, up to 12 months |
| | 5X SiaFind™ Binding Buffer 1 (BA0101) | 100 mL | | 4 to 25°C |
| SK2301B | SiaFind™ α 2,3-Specific Lectenz®, Biotinylated (SP2302B-1MG) | 1 mg, lyophilized | 78,000 | -20°C, up to 12 months |
| | 5X SiaFind™ Binding Buffer 1 (BA0101) | 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 **lectin-like, enzyme-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™ α 2,3-Specific Lectenz® Kits (Cat #SK2301 and SK2301B) contain a sialic acid affinity reagent for the detection, separation, or enrichment of sialoglycans terminating in Neu5Ac α 2,3Gal commonly found in glycoconjugates (glycoproteins, glycolipids, and oligo- or polysaccharides). It has high affinity and specificity towards α 2,3 linked sialic acids on glycans. However, it does not bind effectively to branched sialylated epitopes such as sialyl Lewis A/X. Each kit also includes a 5X binding buffer to ensure maximum reagent specificity and ease of use.

Each **SiaFind™ Lectenz®** has a molecular mass of about 77 kD and works as a monomer without bivalent metal ions. It is 6xHis-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.

Form and Storage

The **SiaFind™ Lectenz®** are supplied lyophilized in a storage buffer (50 mM EPPS, 100 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 $E^{1\%}_{1\text{cm}}$ 12.7. 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 3'-sialyllactose-BSA as positive control.

Prepare 1X SiaFind™ Binding Buffer 1 (SBB1, 10 mM EPPS, 10 mM NaCl, pH 7.5) from the 5X binding buffer (Cat #BA0101). Prepare SBB1 plus 0.1% Tween-20 (SBB1T) for membrane washing.

Prepare SBB1T with 5% globulin free BSA for blocking. Incubate the membrane at room temperature for 1 h with agitation.

Prepare **SiaFind™ Lectenz®** in SBB1T with 0.5% globulin-free BSA: 25 μ g/mL of the native reagent (Cat #SP2302) or 5 μ g/mL of the biotinylated reagent (Cat #SP2302B). Incubate at room temperature for 1 h with agitation. Rinse membrane 3 X 5 min with SBB1T.

Incubate with a 2° probe diluted in SBB1T 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 #SP2302) or 1 μ g/mL of a streptavidin-HRP solution for the biotinylated reagent (Cat #SP2302B). Rinse membrane 3 X 5 min with SBB1T.

Rinse membrane 3 X 5 min with SBB1 before applying HRP chemiluminescent substrate for detection.

Note: *SiaFind™ Lectenz® are sensitive to salt. Titration of NaCl concentration in the binding buffer may be performed. They will work in common Western Blotting buffers, such as PBS or TBS, but the binding signal may be significantly weaker.*