



**LudgerClean S Cartridges**  
for Clean-up of Glycan Samples

**Catalog No**  
LC-S-01

**Lot No**  
BOCH-01

**Application:**  
Purification of glycans after fluorescence labeling.

**Pack Size**  
6 LudgerClean S cartridges

**Kit Storage**  
Store in a dark, dry environment at room temperature.

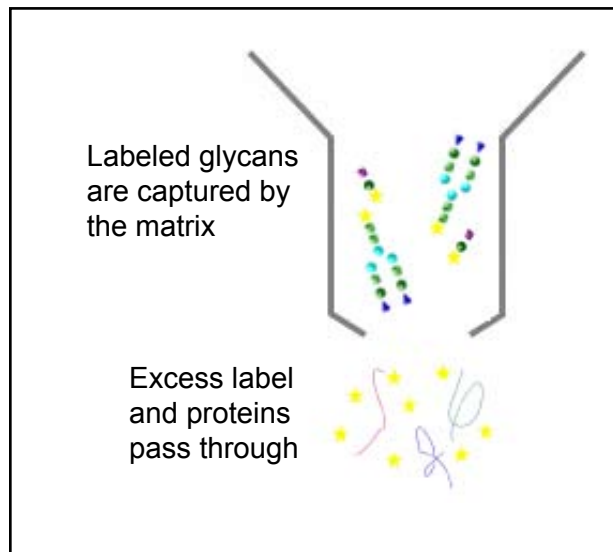
**Amount of Sample**  
up to 20 µg of glycan

**Binding Efficiency**  
>90% for most glycanst

**Binding Selectivity**  
Essentially stoichiometric binding and elution for most complex glycan mixtures.

**Sample Type**  
Recommended for N-linked and O-linked trisaccharides and larger. Monosaccharides or disaccharides are generally bound to weakly for efficient purification.

- Additional Reagents Required**
- Pure water, HPLC grade
  - Acetonitrile, HPLC grade
  - 96% acetonitrile / 4% water (v/v)
  - 30% acetic acid / 70% water (v/v)



**Description**  
S cartridges can be used for the purification of glycans after labeling with QA-Bio's LudgerTag 2-AA (2-amino-benzoic acid) labeling kit (cat number: LT-KAA-A2) and other fluorescent tags.

The procedure is very straightforward. The sample is loaded onto the cartridge's adsorption disc that has been primed with acetonitrile. The glycans adsorb onto the disc matrix while excess dye is removed by washing with an acetonitrile/water mix. The glycans are then desorbed by simply washing with water.

The method can be extended to cleanup of small volumes of glycan samples after a variety of other procedures including reductive amination with other dyes such as 2-aminobenzamide (2-AB, cat number: LT-KAB-A2) and after enzyme digestions.

**LC-S-01 S Cartridges**  
**Specifications - Protocol**

**Glycan Clean-up Protocol**

1. Prepare S cartridges (one cartridge per sample) by washing each with 1 ml water followed by 2 ml 30% acetic acid (aq). After this has drained, wash with 1 ml acetonitrile.

If flow is restricted, e.g. by an air gap, then apply a slight pressure to the top of the cartridge in order to resume normal flow.

2. While the disc is still wet with acetonitrile, spot the sample to be cleaned (maximum volume 15 µl) onto the center of the disc. If your sample has a greater volume, dry it down using a centrifugal evaporator and reconstitute in not more than 15 µl of water.

Make sure that samples in fluorescent labelling mixtures are applied to the disc at room temperature or below. Do not apply warm labelling mixtures to the disc.

If a disc has dried out it should be re-wetted by washing with a further 0.5 ml acetonitrile prior to loading the sample.

When applying the sample to the cartridge, spread the spot over the entire center of the adsorption disc - this will give a more efficient cleanup than a small spot.

3. For maximum recovery, rinse each empty sample vial with 100 µl acetonitrile and apply to the corresponding cartridge disc.
4. Leave for 15 minutes while the glycans adsorb onto the disc.
5. Wash each disc with 1 ml acetonitrile, followed by 5 x 1 ml 96% acetonitrile / 4% water. Discard these washes into a suitable waste container.
6. Place each cartridge over a collection vessel suitable for drying 1.5 ml water and recover the glycans by eluting with 3 x 0.5 ml water, allowing each aliquot to drain before the next is applied.
7. Evaporate to dryness in a rotary or centrifugal evaporator.
8. Redissolve in an appropriate volume of water or solvent then filter before further analysis. Samples should be filtered to at least 0.5 µm.

**References**

1. Bigge, J.C.; Patel, T.P.; Bruce, J.A.; Goulding, P.N.; Charles, S.M.; Parekh, R.B. (1995) 'Non-selective and efficient fluorescent labeling of glycans using 2-aminobenzamide and anthranilic acid'. *Analytical Biochemistry* **230**: 229-238
2. Guile, G.R.; Rudd, P.M.; Wing, D.R.; Prime, S.B.; Dwek, R.A. (1996) 'A rapid and high-resolution high-performance liquid chromatographic method for separating glycan mixtures and analyzing oligosaccharide profiles'. *Analytical Biochemistry* **240**: 210-226
3. Townsend, R.R.; Lipniunas, P.H.; Bigge, C.; Ventom, A.; Parekh, R. (1996) 'Multimode high-performance liquid chromatography of fluorescently labeled oligosaccharides from glycoproteins'. *Analytical Biochemistry* **239**: 200-207
4. Hardy, M.R. (1997) 'Glycan labeling with the fluorophores 2-aminobenzamide and anthranilic acid' in 'Techniques in Glycobiology', edited by Townsend, R.R. and Hotchkiss, A.T.. Marcel Dekker Inc, New York .

**Warranty**

QA-Bio warrants that the above product conforms to the specification described herein. Should the product fail for reasons other than through misuse, QA-Bio will, at its option, replace the product free of charge or refund the purchase price. This warranty is exclusive and QA-Bio makes no other warrants, expressed or implied, including any implied conditions or warranties of merchantability or fitness for any particular purpose.

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