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Product Information

GlycoProfile™ Glycan Clean-up Cartridges

Catalog Number **G4548**Store at Room Temperature

TECHNICAL BULLETIN

Product Description

Glycan analysis has become an increasingly critical aspect of glycomics and proteomics, due to the role of glycoproteins in cell signaling, cell adhesion, immune response, and disease states. Because they tend to have low spectral activity in both UV and visible light, it is often necessary to label glycans in order to enhance detection.

GlycoProfileTM Glycan Clean-Up Cartridges are designed for the post-labeling cleanup of labeled glycans when using the GlycoProfileTM 2-AA or 2-AB Labeling Kits. Glycans adsorb to the membrane while excess dye and other hydrophobic contaminants pass through. Mono- and disaccharides have a weak affinity for the cartridge membrane and tend to bind poorly. Purified glycans are subsequently desorbed and ready for further enzymatic digestion or analysis by a number of methods, including HPLC.

Each cartridge is recommended for single use only.

The maximum sample size is 10 μ L of glycan containing solution and/or 20 μ g of glycan.

Reagents Required but Not Provided

HPLC Grade Water, Product Number 270733 HPLC Grade Acetic Acid Solution (49-51%, Product Number 45754)

HPLC Grade Acetonitrile, Product Number 439134

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

It is recommended that the entire Technical Bulletin be read prior to use.

Storage/Stability

Store the cartridges at room temperature. The cartridges are stable for at least one year at room temperature, as supplied.

Preparation Instructions

- 30% Acetic Acid Solution Prepare a 30% (v/v) acetic acid solution by mixing 6 volumes of the Acetic Acid Solution (Product Number 45754) with 4 volumes of HPLC Grade Water (Product Number 270733). A minimum volume of 5 ml is required per sample.
- 96% Acetonitrile Solution Prepare a 96% (v/v) acetonitrile solution by mixing 96 volumes of HPLC Grade Acetonitrile (Product Number 439134) with 4 volumes of HPLC Grade Water (Product Number 270733).

Note: The acetonitrile concentration is critical. Acetonitrile solutions of <96% will result in sample loss by the inappropriate elution of glycans, particularly low molecular weight glycans.

A minimum volume of 5 mL is required per sample

 HPLC Grade Water - A volume of ~3 mL of HPLC grade water is required per sample.

Procedure

Glycan Adsorption

During the wash procedure the solvent flow may be obstructed by air gaps. Should this occur, apply a slight pressure to the top of the cartridge in order to clear the trapped air and resume normal flow. The liquid samples should be at or below room temperature before spotting onto the cartridge membrane.

- Wash each cartridge with 1 mL of HPLC Grade Water
- 2. Wash each cartridge with 5 mL of 30% Acetic Acid Solution.
- 3. Allow the cartridge to drain completely and then wash with 1 mL of 96% Acetonitrile Solution.
- 4. While the membrane is still wet (following the acetonitrile wash), spot a sample (maximum volume of 10 μ L) onto each cartridge membrane, ensuring that the sample is spread over the entire membrane surface. If the cartridge membrane has dried, apply 0.5 mL of 96% Acetonitrile Solution to moisten.
- 5. The sample vials may be rinsed with 100 μ L of 96% Acetonitrile Solution. Each rinse may then be applied to the appropriate membrane for maximum recovery.
- Allow each cartridge to stand for 15 minutes to ensure complete adsorption of the glycans to the membrane.
- Wash each cartridge with six consecutive 1 mL volumes of 96% Acetonitrile Solution. Allow the cartridge to drain between each 1 mL application. Discard the organic solvent wash appropriately.

Glycan Elution

- Place each cartridge over a collection vessel sufficient for collecting 1.5 mL of water. If aseptic filtration is required, place the cartridge over a 5 mL syringe fitted with a 0.45 μm PTFE filter.
- 2. Elute the glycans by washing with three consecutive 0.5 mL volumes of HPLC Grade Water. Allow each volume to completely drain between elutions.
- 3. Filter the sample as appropriate. Dry the purified glycan samples using a centrifugal evaporator or lyophilizer.
- Re-dissolve the purified glycan samples in a desired volume of solvent or water as appropriate for additional analysis.
- 5. Store samples at -20 °C protected from light in preparation for further downstream analysis.

Related Products

GlycoProfile 2-AA Labeling Kit, Product Number PP0530

GlycoProfile 2-AB Labeling Kit, Product Number PP0520\)

Enzymatic Deglycosylation Kit, Product Number EDEGLY

PNGase F, Product Number P7367 O-Glycosidase, Product Number G1163

References

 Novotny, M.V., and Alley, W.R., Jr., Curr. Opin. Chem. Biol., 17(5), 832-840 (2013).

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