INSTRUCTIONS

PIERCE

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Extracti-Gel® D Detergent Removing Gel

20208 20303 20346

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Product Description

Number	Description
20208	**Extracti-Gel® D Detergent Removing Gel,** *10 ml gel*
20303	**Extracti-Gel® D Detergent Removing Gel,** *100 ml gel*
20346	**Extracti-Gel® D AffinityPak™ Columns,** *5 x 1 ml*

Introduction

Extracti-Gel® D Detergent Removing Gel has low non-specific binding characteristics. However, since this gel has an exclusion limit of 10,000 MW, components of lower molecular weight will be able to enter the pores and interact with the affinity ligand. This may result in the loss of small molecules such as peptides. We recommend that macromolecules from which detergents are to be removed have a molecular weight in excess of 10,000. Additionally, solutions that are extremely dilute with respect to the concentration of a macromolecule (i.e. 50 µg/ml or less) may result in a severe yield loss due to non-specific binding regardless of the type of chromatographic support used. In such cases, we recommend the inclusion of a carrier molecule such Bovine Serum Albumin (0.1%) in the sample to mask any of these sites so that high recoveries may be obtained. Extremely hydrophilic detergents may not bind to this gel as well as hydrophobic ones.

Using the Extracti-Gel® Packed Column

1. Allow buffers and gel to come to room temperature. The column may be run at 4°C, however flow rates will be slow and the sample may have to be passed through the gel several times to remove all of the detergent.

   Note: The procedure may be performed at 4°C when removing Surfact-Amps™ Triton® X-114 because this detergent tends to be cloudy at room temperature.

2. Wash and equilibrate the column with a suitable, degassed buffer.
3. Move column to a new test tube.
4. Apply the sample to the column. The proteins will be eluted in the void volume (0.7 -1.0 ml/ml of gel).

   Note: Because the exclusion limit of the gel is 10,000 MW, components that have molecular weights lower than this value will be able to enter the pores and interact with the affinity ligand.

5. Apply buffer to the column to allow the sample to pass through the gel.
6. Collect the eluted sample. The detergent should be eliminated from the sample if the maximum capacity of the gel is not reached.

Extracti-Gel® D Detergent Removing Gel Regeneration Scheme

The support may be regenerated by washing the gel according to the following protocol:

1. Wash the gel with one bed volume of distilled/deionized water followed by serially washing with increasing concentrations of ethanol in water, followed by one bed volume of absolute ethanol.
2. Wash with 2 bed volumes of butanol followed by one bed volume of ethanol.
3. Serially wash with ethanol and decreasing concentrations of ethanol in distilled/deionized water followed by one bed volume of distilled/deionized water.

4. Equilibrate with the buffer from which detergent is to be removed.

This gel has been successfully regenerated three times.

Note: Do not let the gel stay immersed in solvent.

Detergent binding capacity of Extracti-Gel® detergent removing gel:

Detergent	Prod. No.	Capacity (mg/ml gel)	Binding buffer
CHAPS	28300	50	50 mM Tris, pH 9.0
SDS	28312	80	50 mM Tris, pH 9.0
Triton X-100	28314	57	100 mM Phosphate, pH 7.0
Triton X-114	28322	85	100 mM Phosphate, pH 7.0
Brij 35	28316	80	100 mM Phosphate, pH 7.0
Tween 20	28320	74	100 mM Phosphate, pH 7.0
Lubrol PX	28322	106	100 mM Phosphate, pH 7.0
NP-40	28324	75	100 mM Phosphate, pH 7.0

These binding studies were performed at room temperature, except for Triton® X-114 which was done at 4°C due to the low cloud point of that detergent.

The Pierce detergents are provided as 10% solutions in sealed ampules. Our detergents are guaranteed low in peroxide and carbonyl content.

References

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3. Sibley, D.R., Strasser, R.H., Benovic, J.L., Daniel, K. and Lefkowitz, R.J. (1986) Phosphorylation/Dephosphorylation of the ß-Adrenergic Receptor Regulates its Functional Coupling to Adenylate Cyclase and Subcellular Distribution, *Proc. Natl. Acad. Sci. U.S.A.* **83,** 9408-9412.
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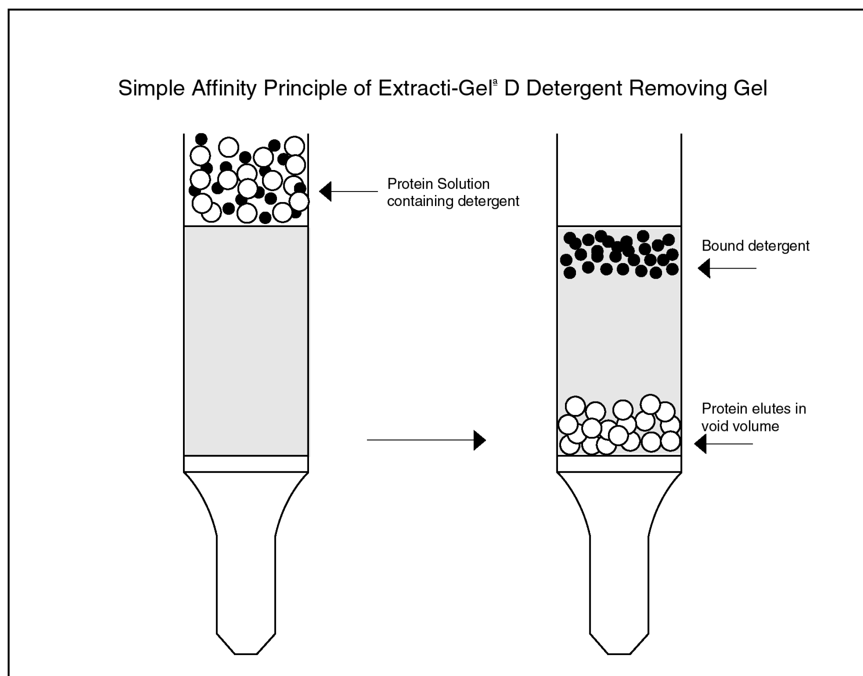

Simple Affinity Principle of Extracti-Gel® D Detergent Removing Gel