# **INSTRUCTIONS**



# Immobilized *p*-Aminophenyl Phosphoryl Choline Gel

20307

Number Description

20307 Immobilized *p*-Aminophenyl Phosphoryl Choline Gel, 5mL

Support: Crosslinked 6% beaded agarose

Binding Capacity: ≥ 3mg of human C-reactive protein per milliliter of gel

**Storage:** Upon receipt store at 4°C. Product is shipped at ambient temperature.

# Introduction

Within the ascites fluid of humans and rabbits exist several different types of immunological proteins that provide aid during an inflammatory response. One of these proteins is the C-reactive protein that has been linked to several biological functions including activation of the classical complement pathway, enhancement of phagocytosis and interaction with certain subpopulations of T-lymphocytes. In the late 1970s, Volanakis and colleagues discovered a method of isolating and studying C-reactive protein using the protein's affinity for phosphoryl choline. Thermo Scientific Immobilized p-Aminophenyl Phosphoryl Choline Gel enables purification of C-reactive protein in a convenient column format that is quick and easy to use.

# **Additional Materials Required**

- Sample purified from ascites fluid by centrifugation and filtration
- Binding Buffer: 0.1M Tris or Borate Buffer, 0.1-0.2M NaCl, 1-2mM CaCl<sub>2</sub>; pH 8-8.5
- Elution Buffer: 0.1M Tris or Borate Buffer, 0.1-0.2M NaCl, 2mM EDTA; pH 8-8.5
- Storage Buffer: 0.01M sodium phosphate, 0.09M NaCl

## **Procedure for C-reactive Protein Purification**

- 1. Prepare a 5mL column by packing gel into a disposable column.
- 2. Equilibrate column by applying two column volumes of Binding Buffer.
- 3. Add 2-3mL of purified sample to column.
- 4. Incubate for 1 hour at room temperature.
- 5. Wash with 5 column volumes of Binding Buffer.
- 6. Elute bound protein with Elution Buffer. Collect fractions in 0.5mL aliquots.
- 7. After elution, micro-concentrate the protein and dialyze into the Storage Buffer.

### **Cited Reference**

1. Volanakis, J.E. *et al.* (1978). C-reactive protein: Purification by affinity chromatography and physicochemical characterization. *J Immunol Meth* 23:285-95



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