

NOTE NO. 21 - June 30 1987

## MOLECULAR WEIGHT STANDARDS ON PINKERTON ISRP

**Analytes:** Chromatogram: #I: Aprotinin (MW=6,500), #II: Cytochrome C (MW=12,400), #III: Carbonic Anhydrase (MW=29,000), #IV: Globulin: (MW=55,000), #V: BSA (MW=66,000), #VI: Blue Dextran (MW=2,000,000)

**Sample Matrix:** 2.5 mg/ml in 0.1 M  $\text{KH}_2\text{PO}_4$

**Sample Size:** 20 microliters

**Column:** Guard: 5 micron GFF ISRP, 1 cm x 3.0 mm ID

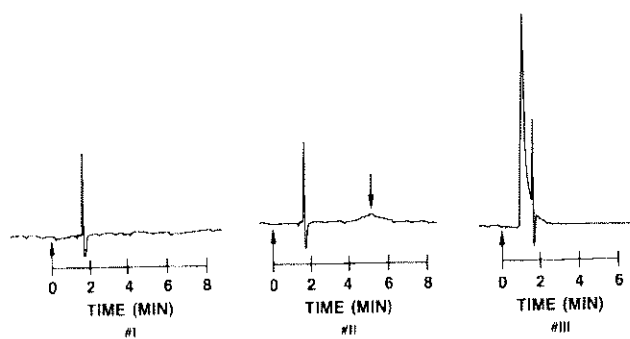
**Regis Product Number:** 731440

**Analytical:** 5 micron GFF ISRP, 15 cm x 4.6 mm ID

**Regis Product Number:** 731451

**Mobile Phase:** 90% 0.1 M  $\text{KH}_2\text{PO}_4$  (pH 6.8), 10% Acetonitrile (HPLC grade reagents)

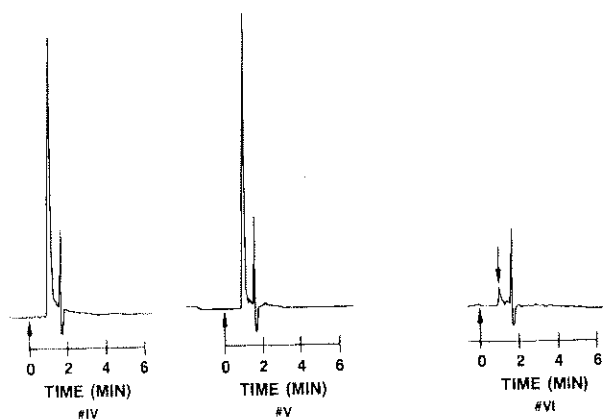
**Discussion:** The size-excluded proteins are not retained, as ISRP theory suggests (indeed, they are detected earlier than the "dead-volume" signal). These 5.2-nm pores apparently exclude proteins heavier than about 20,000-25,000 daltons--for instance, the 29,000-dalton Carbonic Anhydrase. The 12,400-dalton Cytochrome C and 6,500-dalton Aprotinin, however, enter the pores and are retained (the Aprotinin, retained so long it could not be detected).



**Detection:** 254 nm, 0.16 AUFS

**Flow Rate:** 1.0 ml/min

**Note:** Sample filtered through 0.2  $\mu\text{m}$  membrane prior to direct injection



**J.D. Rateike, Regis Chemical Company,  
8210 Austin Avenue, Morton Grove, IL 60053**