IEC
TSKgel BioAssist SERIES

APPLICATIONS WITH TSKgel BioAsisst ANION EXCHANGE COLUMNS

Comparison of Dynamic Binding Capacity

Table 2 shows typical dynamic binding capacities on Bio-Assist Q relative to competitive products.

TABLE II

COMPARISON OF DYNAMIC BINDING CAPACITIES

	Binding capacity (mg/mL)			
Protein	BioAssist Q	SuperQ -5PW	Conv. Q type prod. A	Conv. Q type prod. B
Thyroglobulin	77.4	22.9	20.2	1.8
Monoclonal IgG_1	57.8	43.3	46.7	47.7
Human Serum Albumin	83.1	78.9	48.2	48.8
Trypsin Inhibitor	84.3	92.8	51.8	57.8

Columns: TSKgel BioAssist Q and TSKgel SuperQ-5PW (4.6 mm ID x 1 cm); Conventional Q type product A and B (4.6 mm ID x 1 cm)
Solvent: 20 mmol/L Tris-HCl buffer, pH 8.0;
Flow rate: 0.38 mL/min; Det.: UV @ 280 nm
*Capacity was determined at 10% height of the breakthrough curve; UV 280 nm.

APPLICATIONS WITH TSKgel BioAssist CATION EXCHANGE COLUMNS

Bromelain Anaylsis on TSKgel BioAssist S and Competitor S Column

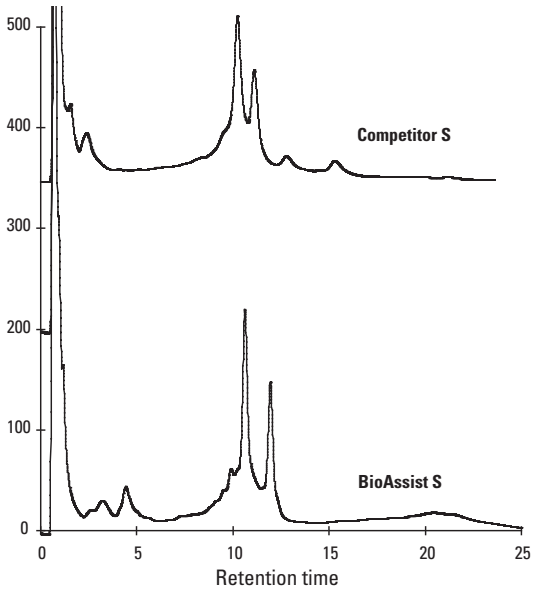
Figure 12 shows the analysis of bromelain, a proteolytic enzyme that is used as a nutritional supplement. Bromelain is a basic glycoprotein with a MW of 33 kDa and a pI of 9.55.

Analysis of IgM

IgM is known to possess unique and beneficial characteristics relative to other immunoglobulin classes; it is a large molecule comprised of five IgG subunits, resulting in a relatively unstable and difficult to purify protein. Unlike single chain antibodies, IgM cannot be purified by Protein A (affinity material commonly used for its high binding capacity and excellent selectivity for antibodies) due to steric hindrance. Alternative affinity methods have been developed with thiophillic absorbents but these methods often result in low binding capacity. An alternative purification method of IgM by ion exchange chromatography using a TSKgel BioAssist S column was developed. Figure 13 shows the baseline separation of IgM from other contaminants using a 0.3 mol/L NaCl step gradient after elution of albumin.

FIGURE 12

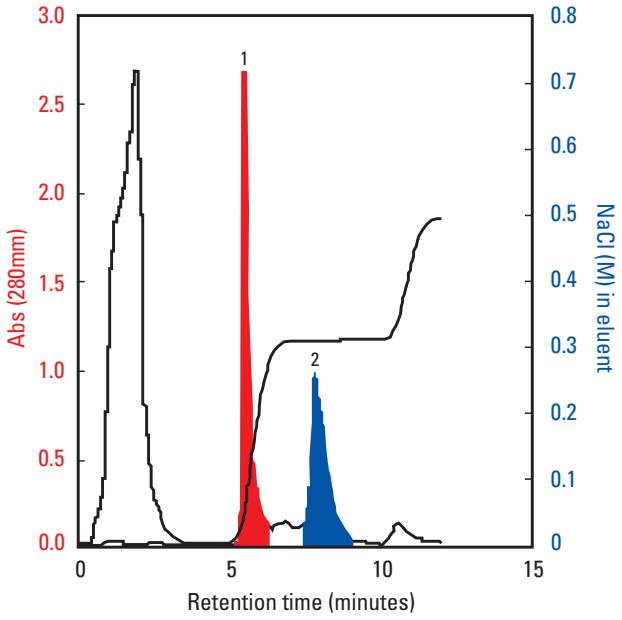
BROMELAIN ANALYSIS ON TSKgel BioAssist S & COMPETITOR S COLUMNS



Columns: TSKgel BioAssist S, 4.6 mm ID x 5 cm L, PEEK
Competitor S 5 mm ID x 5 cm L; Elution: 20 min (TSKgel) or 30 min (Competitor S) linear gradient of NaCl from 0 to 0.5 mol/L in 20 mmol/L sodium phosphate buffer, pH 7.0; Flow rate: 0.8 mL/min for TSKgel; 1.0 mL/min for Competitor S
Detection: UV @ 280 nm; Temperature: 25°C;
Sample: crude bromelain (C4882, Sigma), 1 mg in 100 µL

FIGURE 13

ANALYSIS OF IgM



Column: TSKgel BioAssist S, 7 µm, 4.6 mm ID x 5 cm L;
Mobile phase: 20 mmol/L sodium phosphate buffer, pH 6.0;
Gradient: 0 mol/L - 0.3 mol/L NaCl (5 min), 0.3 mol/L - 0.5 mol/L NaCl (10 min);
Flow rate: 1 mL/min; Detection: UV @ 280 nm; Sample: 500 µL of 9.5 mg/mL IgM in mouse ascites fluid; shaded peaks represent albumin and IgM respectively