Essentials in biore

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FASEB

Poster Presentation

Isolation of Prostatic Acid Phosphatase on a New Preparative Ion Exchanger

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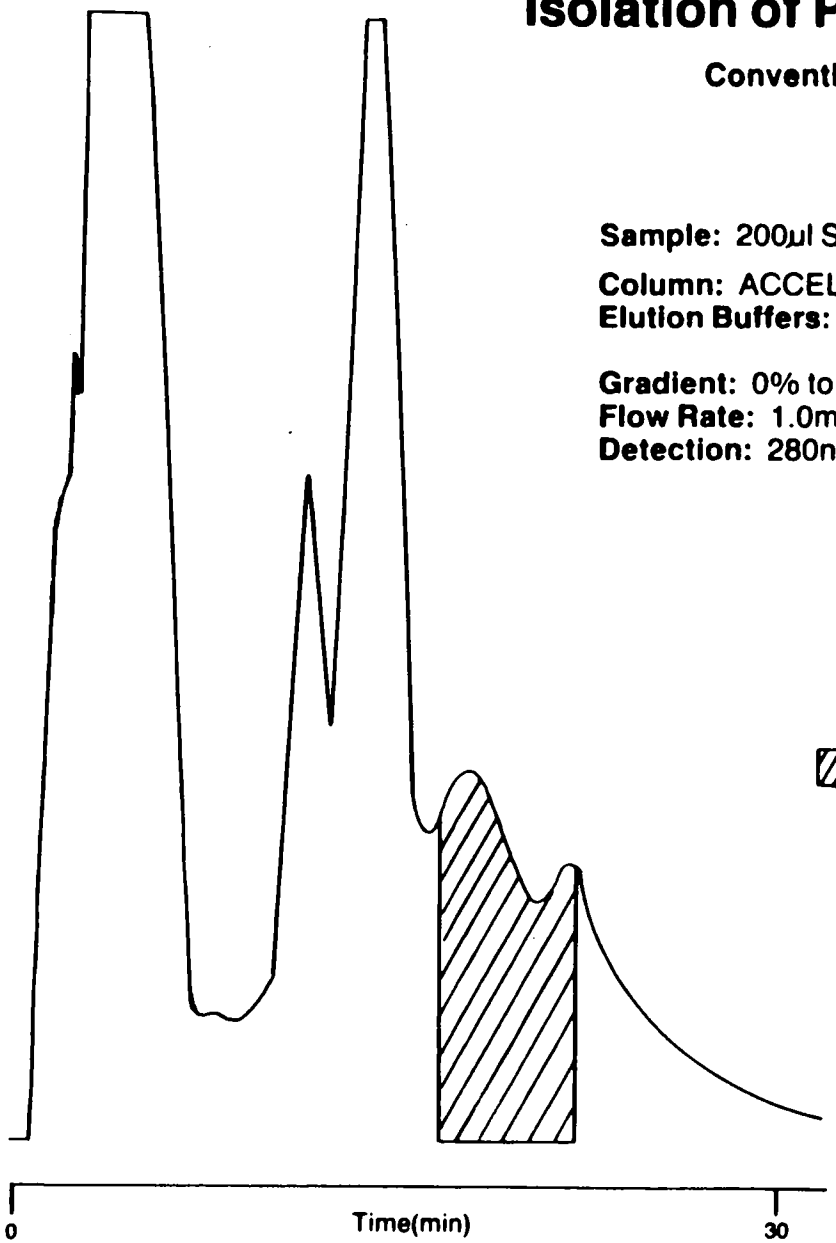
Prostatic Acid Phosphatase

M.W.	:	102,000 Daltons
Source	:	Seminal Plasma
Characteristics	:	Dimeric, Glycosylated with 11 Known Subforms

Isolation of Prostatic Acid Phosphatase

Conventional DEAE-Cellulose Conditions

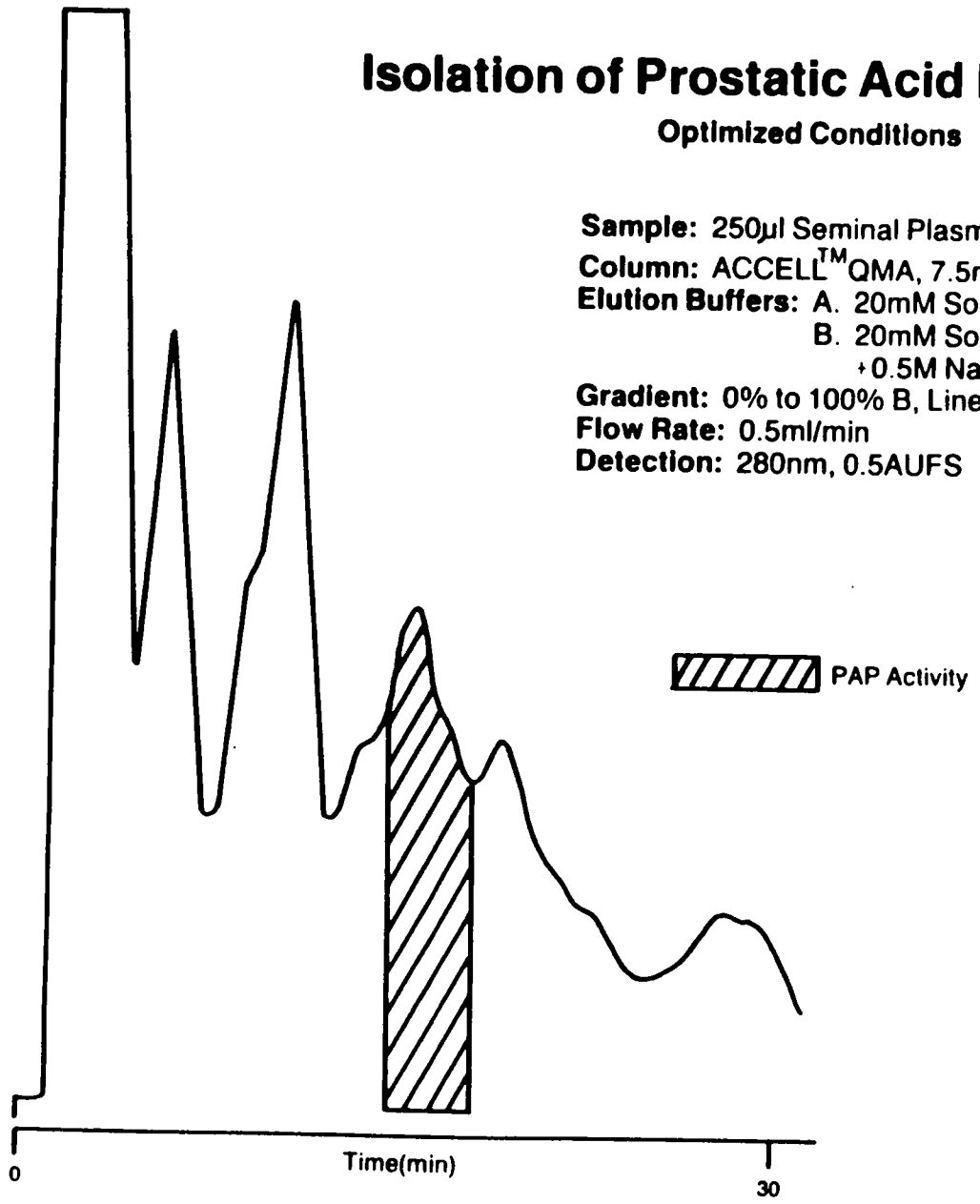
Sample: 200µl Seminal Plasma
Column: ACCELL™ QMA, 7.5mm x 7.5cm
Elution Buffers: A. 20mM TRIS, pH 8.0
B. 20mM TRIS, pH 8.0 + 1.0M NaCl
Gradient: 0% to 50%B, Linear in 30min
Flow Rate: 1.0ml/min
Detection: 280nm, 0.5AUFS



Isolation of Prostatic Acid Phosphatase

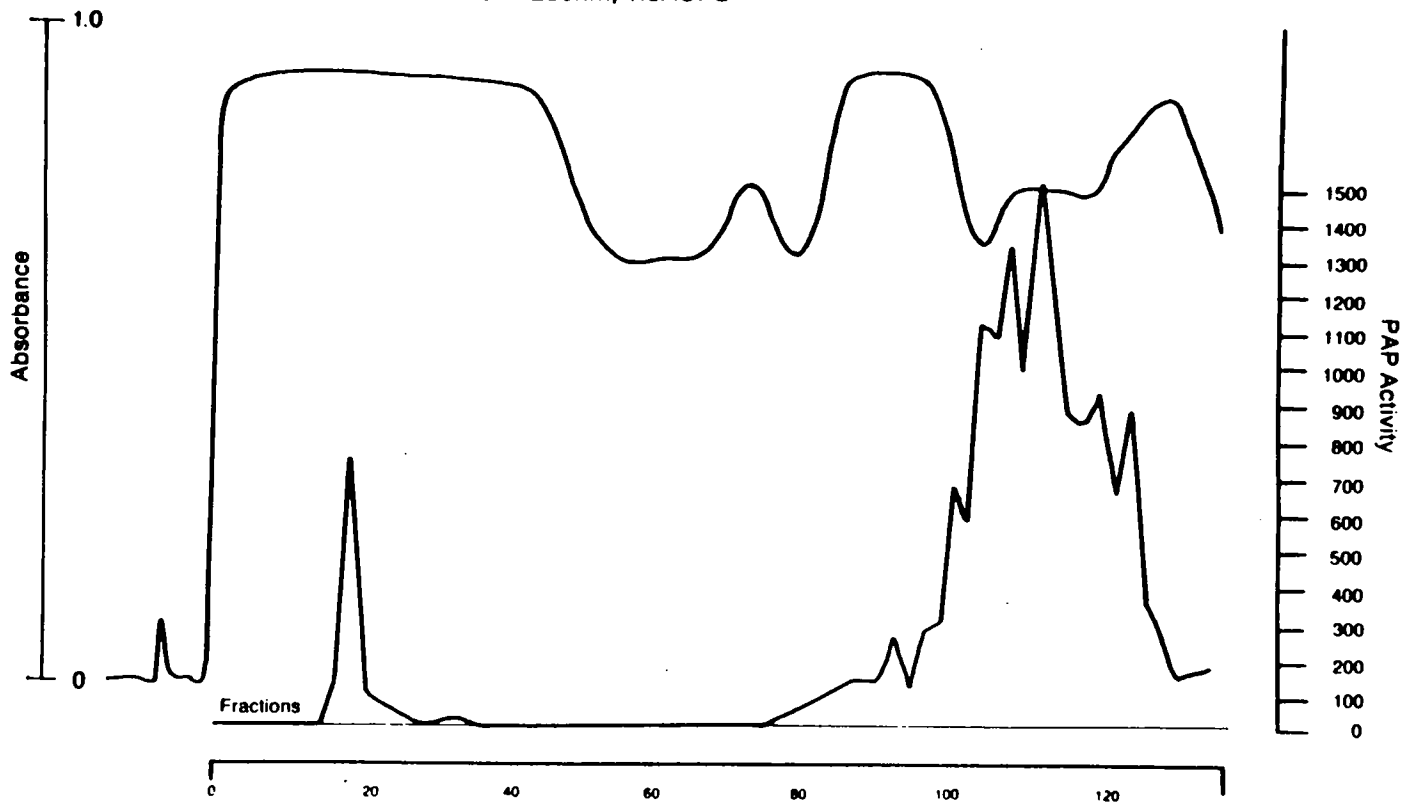
Optimized Conditions

Sample: 250µl Seminal Plasma
Column: ACCELL™QMA, 7.5mm x 7.5cm
Elution Buffers: A. 20mM Sodium Phosphate, pH 7.0
B. 20mM Sodium Phosphate, pH 7.0 +0.5M NaCl
Gradient: 0% to 100% B, Linear in 30min
Flow Rate: 0.5ml/min
Detection: 280nm, 0.5AUFS



Preparative Isolation of Prostatic Acid Phosphatase

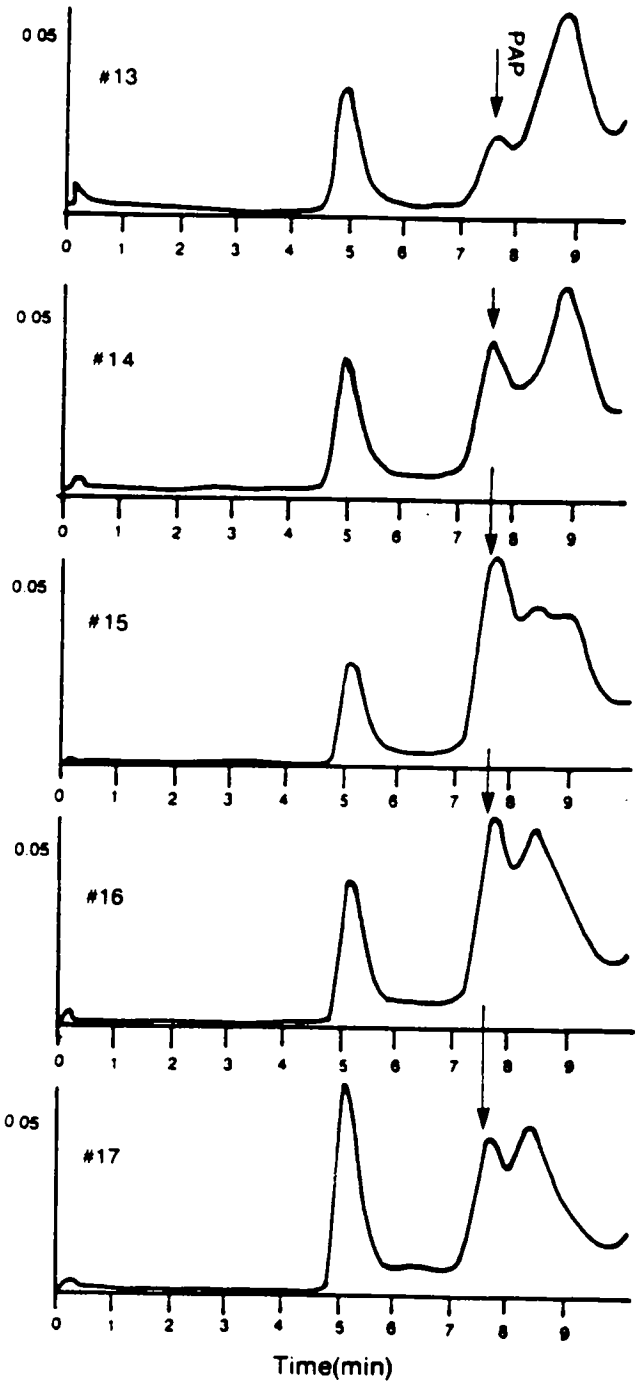
Sample: 20ml Seminal Plasma Diluted
With 20ml 20mM Na_2HPO_4, pH7.0
Column: ACCELL™ QMA, 25mm x 25cm
Elution Buffers: A. 20mM Sodium Phosphate, pH 7.0
B. 20mM Sodium Phosphate, pH 7.0
+0.5M NaCl
Gradient: 0% to 80% B, Linear in 165min
Flow Rate: 2.0ml/min
Detection: 280nm, 1.0AUFS



Isolation of Prostatic Acid Phosphatase

Rechromatography of Active Fractions

Samples: PAP-Containing Fractions From ACCELL™ QMA Isolation in pH 7.0 Phosphate Buffer
Column: PROTEIN-PAK™ 300sw, 7.8mm x 30cm
Elution Buffer: 0.1M Sodium Phosphate, pH 6.8
Flow Rate: 1.0 ml/min
Detection: 214nm, 0.05AUFS



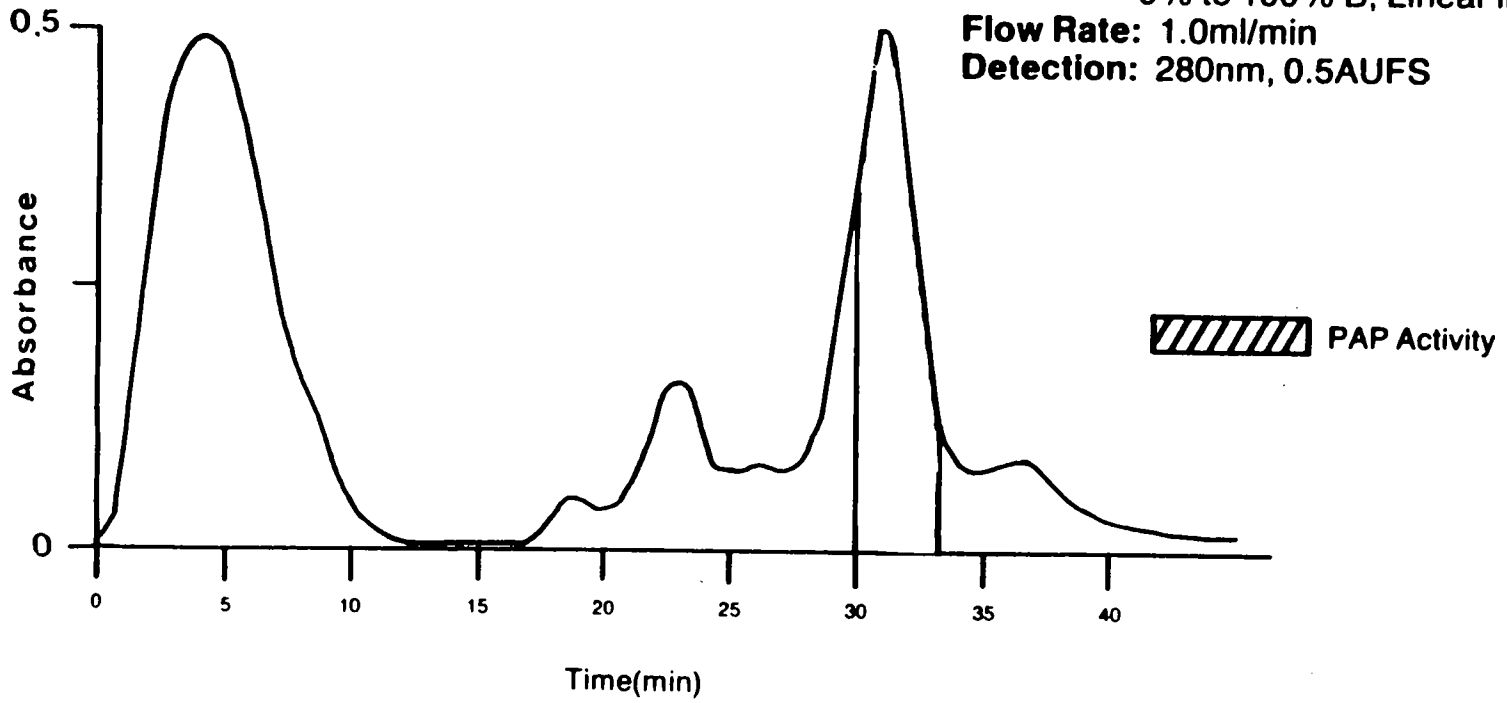
Semi-Preparative Isolation of Prostatic Acid Phosphatase

Sample: 5ml PAP Fraction Pool From Preparative Anion Exchange Isolation

Column: ACCELL™ CM, 7.5mm x 7.5cm

Elution Buffers: A. 20mM Sodium Acetate, pH 4.0
B. 20mM Sodium Acetate, pH 4.0 + 1.0M NaCl

Gradient: 100% A, Isocratic For 10min
0% to 100% B, Linear in 15min

Flow Rate: 1.0ml/min

Detection: 280nm, 0.5AUFS