

1. Background

Risks associated with transmission of pathogenic infectivity through blood transfusions remain high despite efforts in the development of sensitive assays to screen potential donors. These risks can be curtailed through the removal of the pathogenic contaminants via adsorption to affinity ligands immobilized on chromatographic resins.

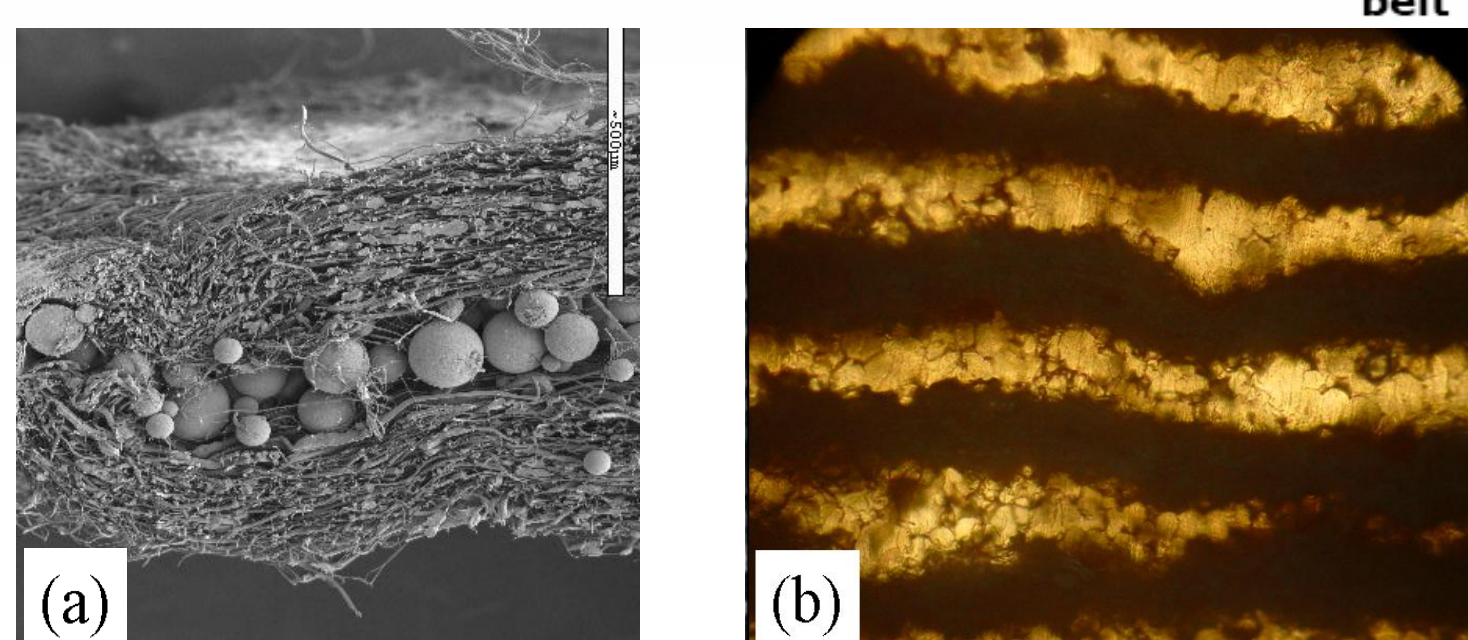
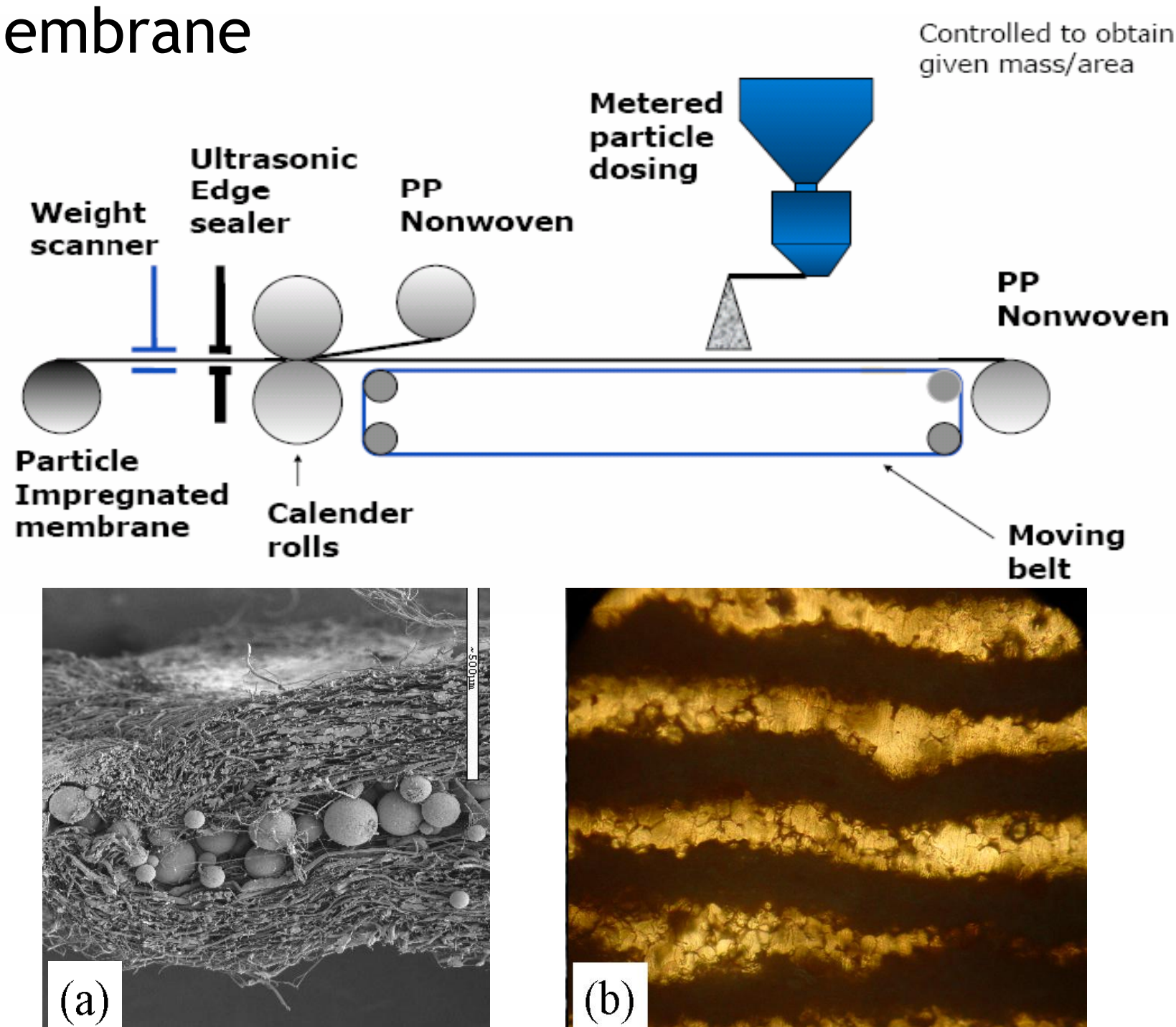
However, the small interstitial hydraulic radii of typical packed-bed columns can preclude separations from sources containing large particles, such as red blood cells. One approach to circumvent this limitation is to entrap chromatographic resin particles in a nonwoven membrane support, allowing larger particles to pass unimpeded while maintaining high surface contact with the chromatographic resin.

2. Methods

Particle-impregnated membranes (PIMs) were produced by spreading an even layer of a polymeric resin between two layers of nonwoven polypropylene membrane. The material was bonded together by calendaring, and cut into circles, which were then stacked together and packed into columns. Challenge solutions (human plasma and red blood cell concentrate (RBC)) were passed through the columns at different flow rates. Interstitial porosity was determined using first absolute moment analysis using pulse experiments.

3. PIM Production

Particle-impregnated membrane was produced as below, resulting in a layer of resin between two sheets of nonwoven membrane

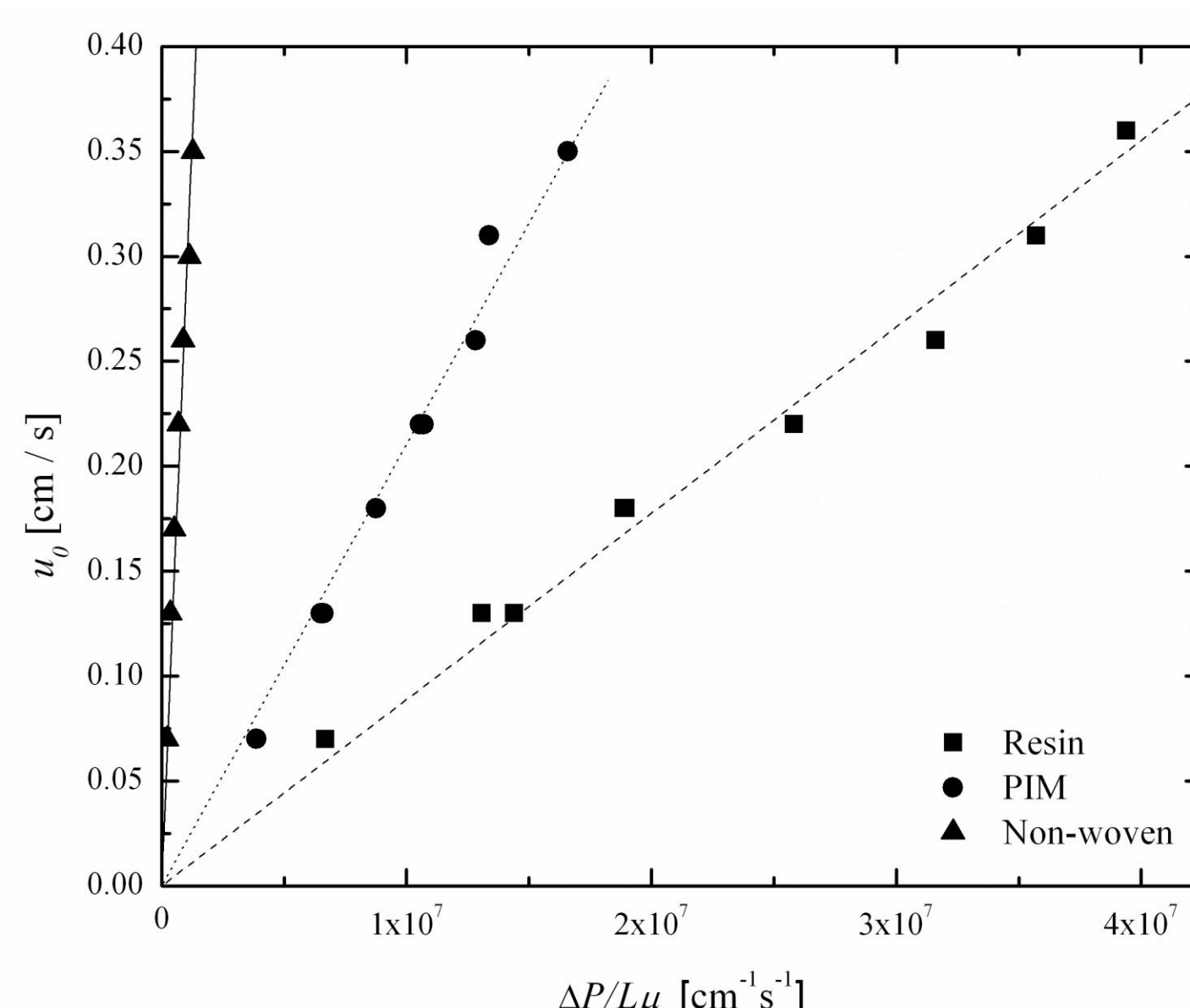


(a) Scanning electron microscopy of PIM materials
 (b) Inverted microscopy of membrane. The dark areas are the nonwoven membrane and the light areas are the resin layers

4. Porosity

Interstitial bed porosities and permeability coefficients measured for a column packed with PIM, resin, and nonwoven membrane

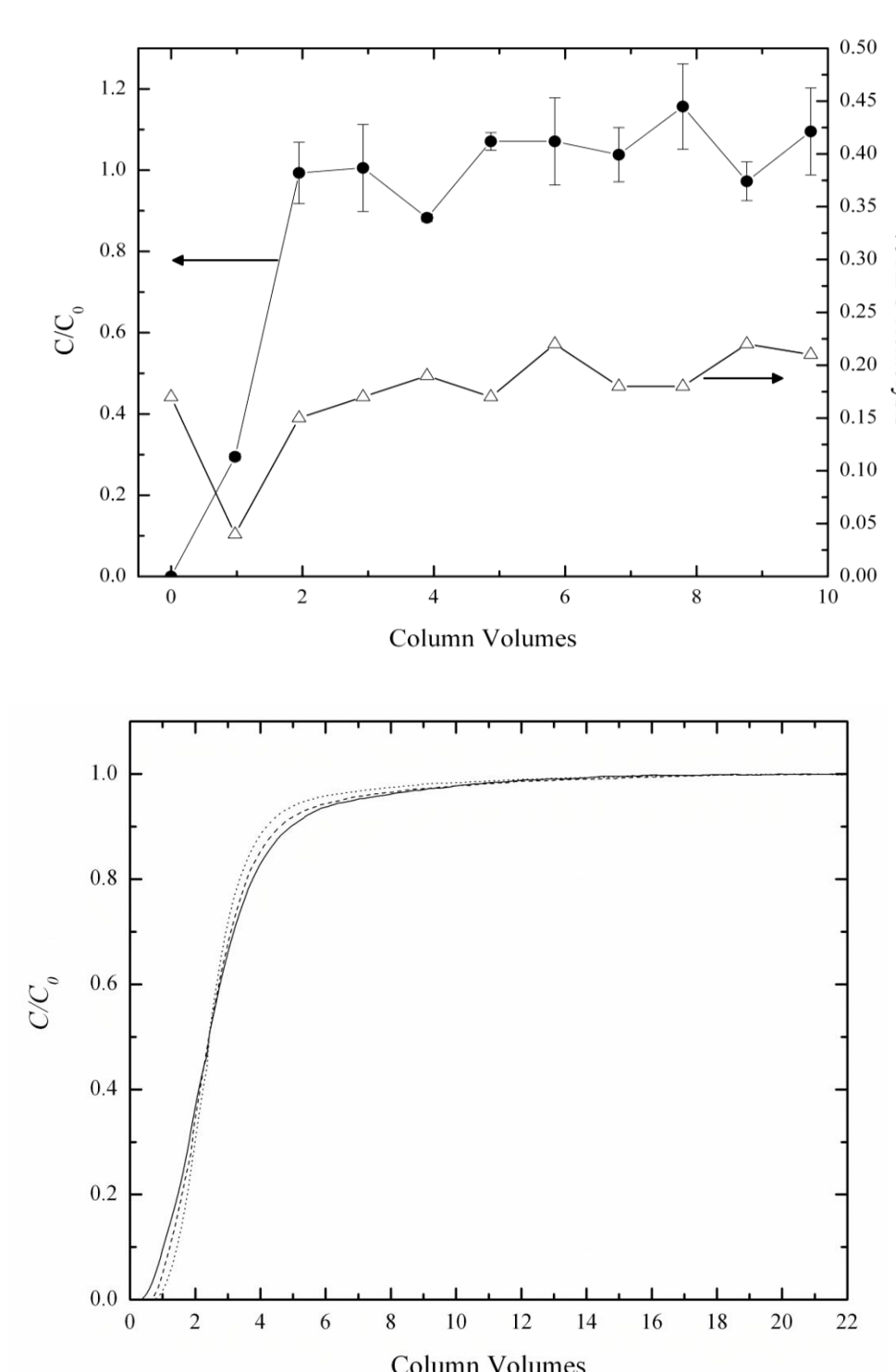
Material	Porosity	κ [cm ²]
Nonwoven membrane	0.75	2.9×10^{-7}
PIM	0.49	2.1×10^{-8}
Resin	0.32	8.79×10^{-9}



Pressure drop-flow rate data for columns packed with nonwoven membrane, PIM, and resin. Permeability coefficients were determined through linear regression

Particle-Impregnated Membrane has higher interstitial bed porosity than resin packed bed

5. Performance with Particulate-Containing Suspensions



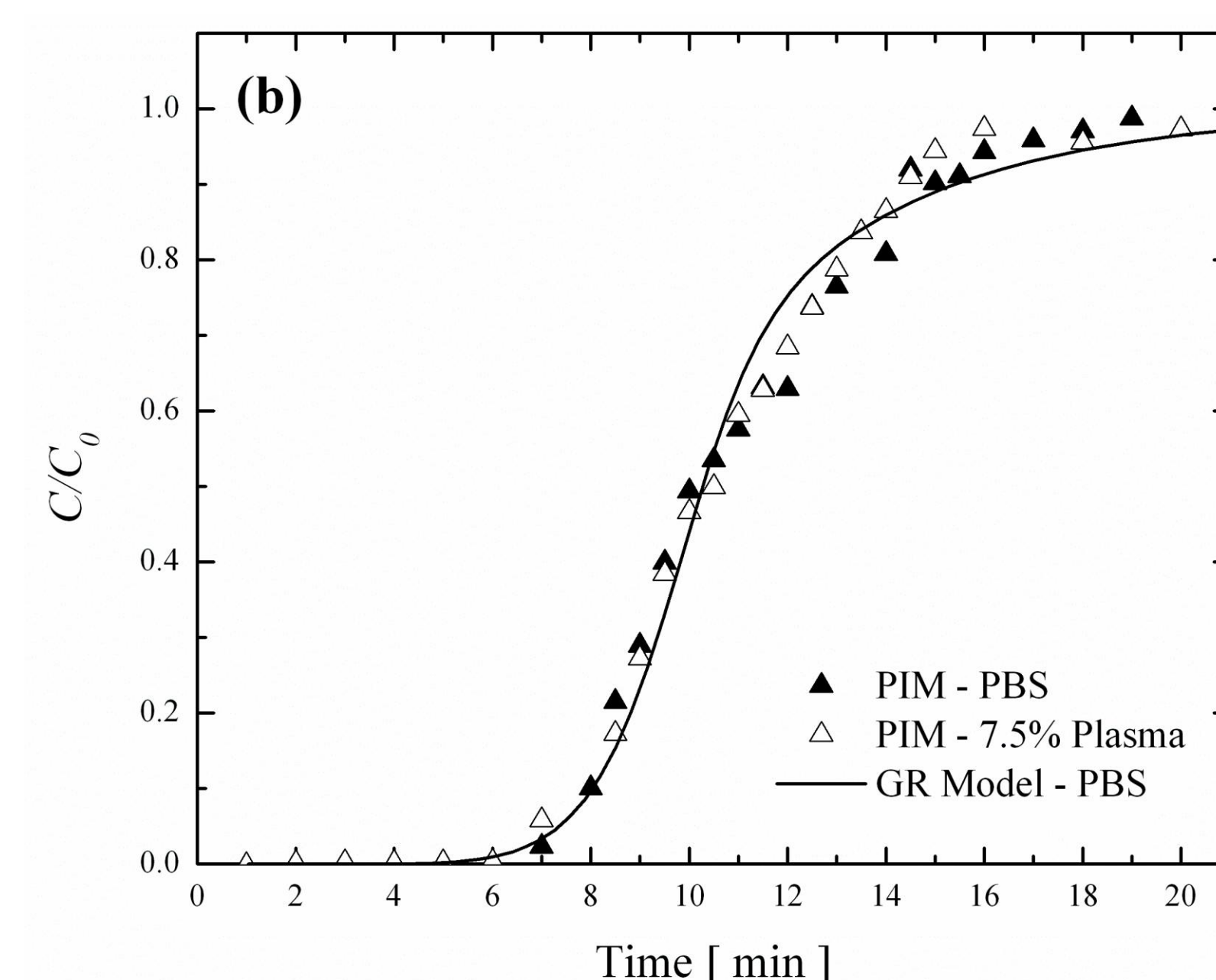
Breakthrough (solid symbols) and resulting haemolysis (open symbols) of RBC fed to a PIM packed column at a linear flow velocity of 0.014 cm/s

Particle-Impregnated Membrane does not damage red blood cells

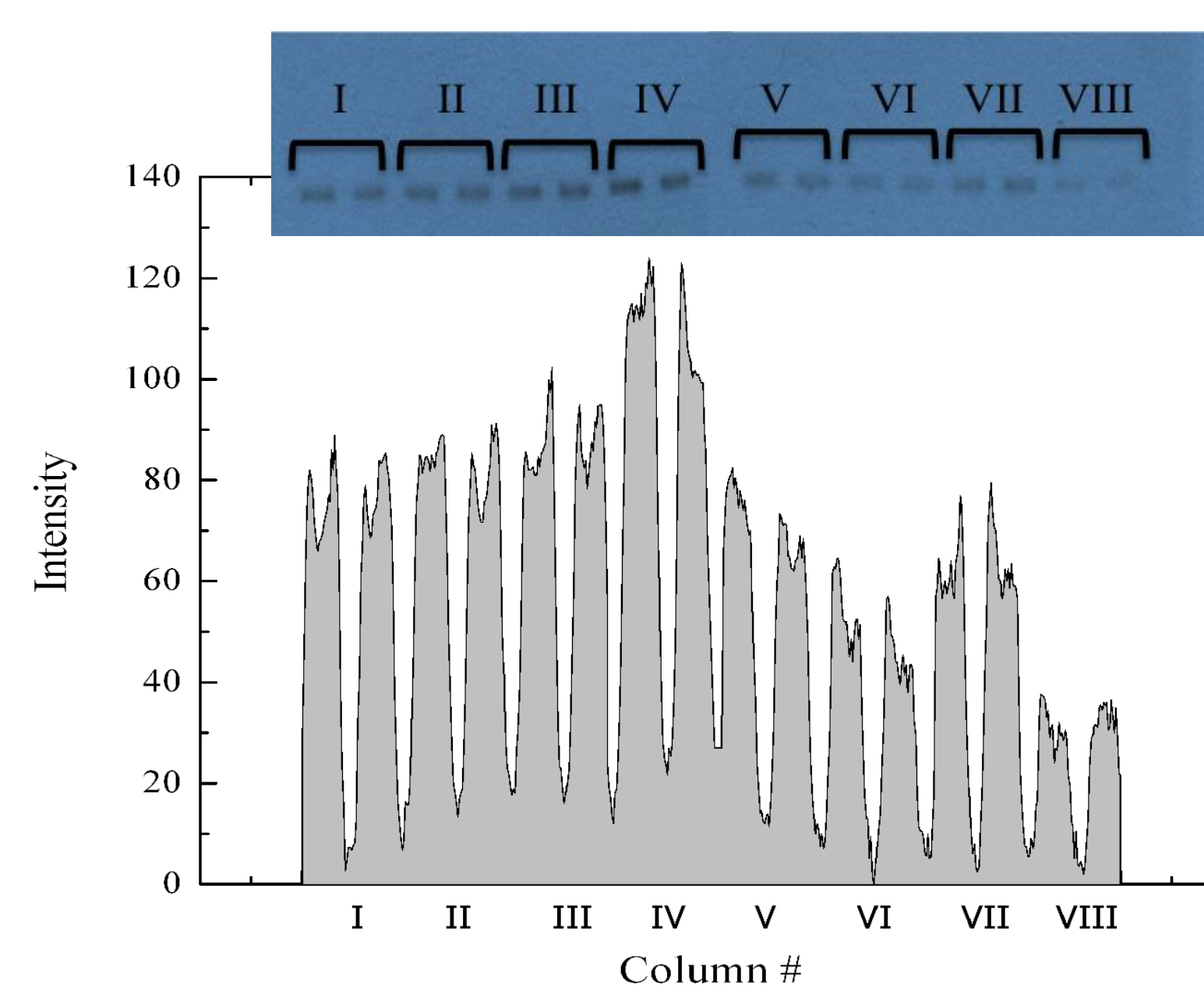
Breakthrough from three different 30-column volume injections of whole cell *E. coli* fermentation suspension to a PIM packed column at a linear flow velocity of 0.014 cm/s.

Particle-Impregnated Membrane retains a negligible amount of *E. coli* cells

6. Binding of Prions in Plasma and RBC



- ❖ Breakthrough of PrP from a 40-layer column of PIM challenged with 1% normal hamster brain homogenate spiked into 7.5% human plasma
- ❖ PrP concentration was monitored by ELISA
- ❖ Spike used is 7 to 8 orders of magnitude larger than expected contamination in plasma



- ❖ Eight PIM columns in series challenged with 0.05% normal hamster brain homogenate spiked into human leukoreduced red blood cell concentrate
- ❖ First 4 columns were saturated
- ❖ Spike used is 5 to 6 orders of magnitude larger than expected contamination in blood products
- ❖ Western blot shows bound fraction

Particle-Impregnated Membrane removes prion protein from human plasma and red blood cell concentrate