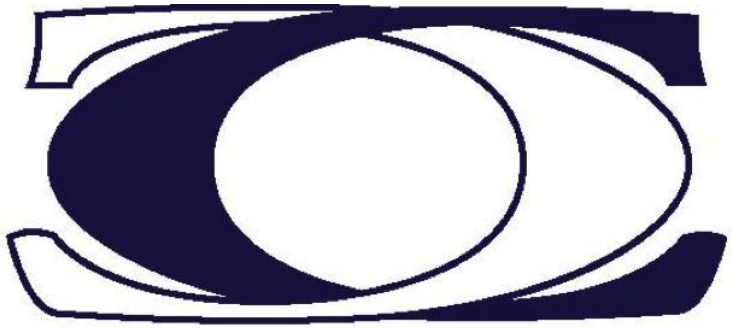


CHIRAL TECHNOLOGIES INC

DAICEL GROUP

move easily
move reliably
move quickly

move ahead

Introduction

The investigation of many different mobile phase components is of critical importance in optimization of preparative chromatography due to their strong influence on the separation and on the production rate of the process.

In enantioselective chromatography, certain solvents such as ethyl acetate and THF have not been available for use with conventionally coated polysaccharide CSPs. Although they are good solvents for the samples, they are also excellent solvents for the chiral polymers. The development of immobilized polysaccharide-based chiral stationary phases (CSPs) such as CHIRALPAK® IA™ has allowed the study of these previously "forbidden" mobile phases.

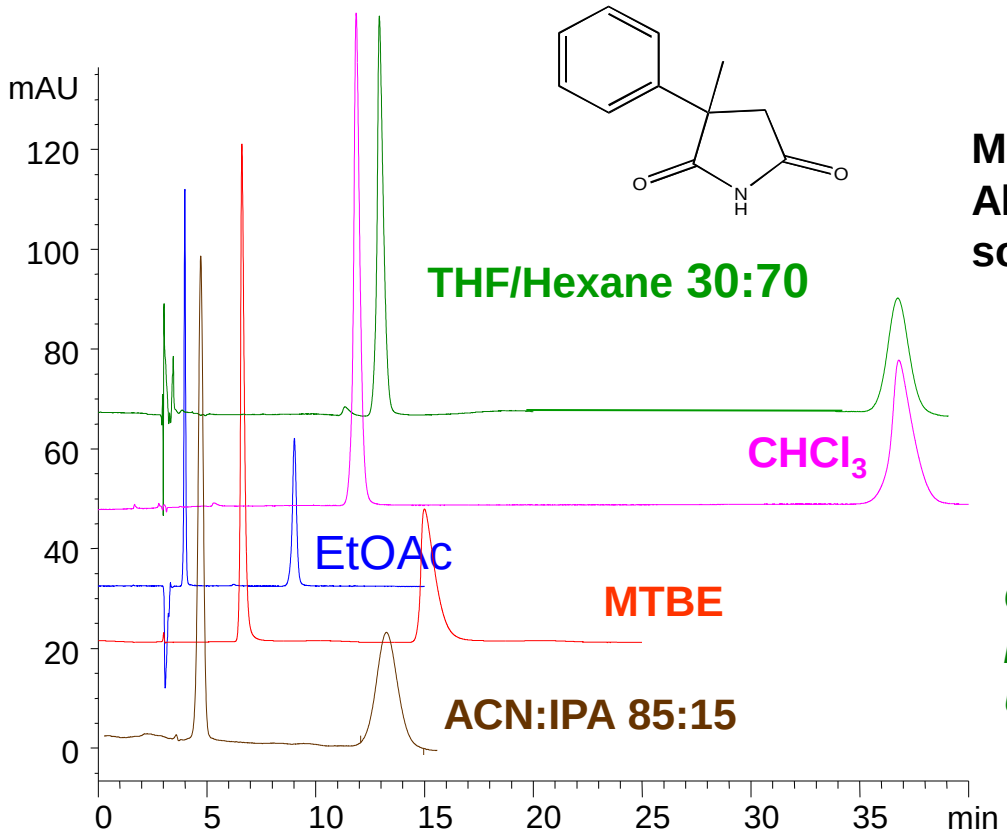
Use of such solvents can influence profoundly the adsorption isotherms for the separations, leading to major improvements in production rate. This poster concludes an ongoing study of solvent effects on SMB separations by presenting the SMB separation of the enantiomers of 1-methyl-1-phenylsuccinimide (MPS) using THF- based mobile phase.

Solvent selection in Preparative LC

- **High selectivity**
- **Short retention times**
- **High sample solubility***
- **Easy to evaporate**
- **Promote high loading***
  - **Need to avoid bi-Langmuir isotherms**
  - **Need high saturation capacity**

Not always easy for chiral separations!

Case Study – α-Methyl-α-Phenylsuccinimide

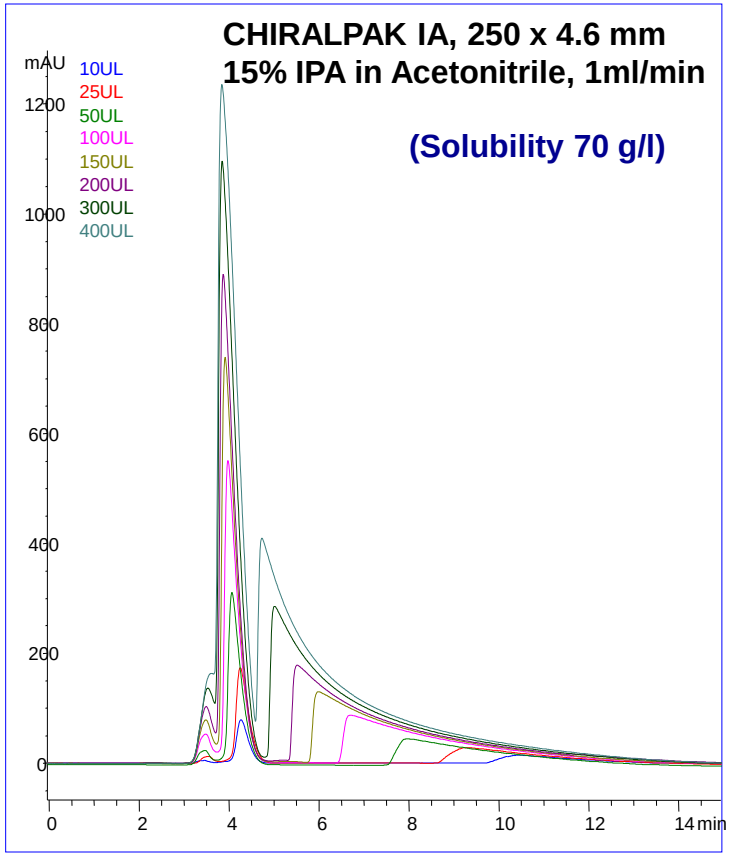


Multiple separation opportunities
Also separates with conventional solvents on CHIRALPAK IA

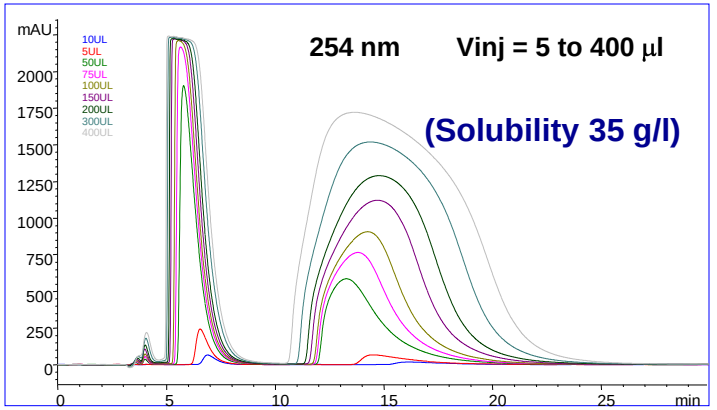
CHIRALPAK IA, 250 x 4.6 mm
Flow rate 1 ml/min
UV detection 254 nm

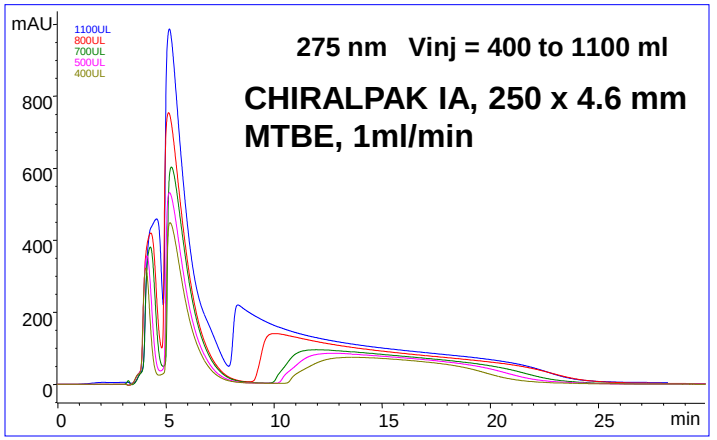
Previously Reported Results

Loading Studies – ACN/IPA; MTBE



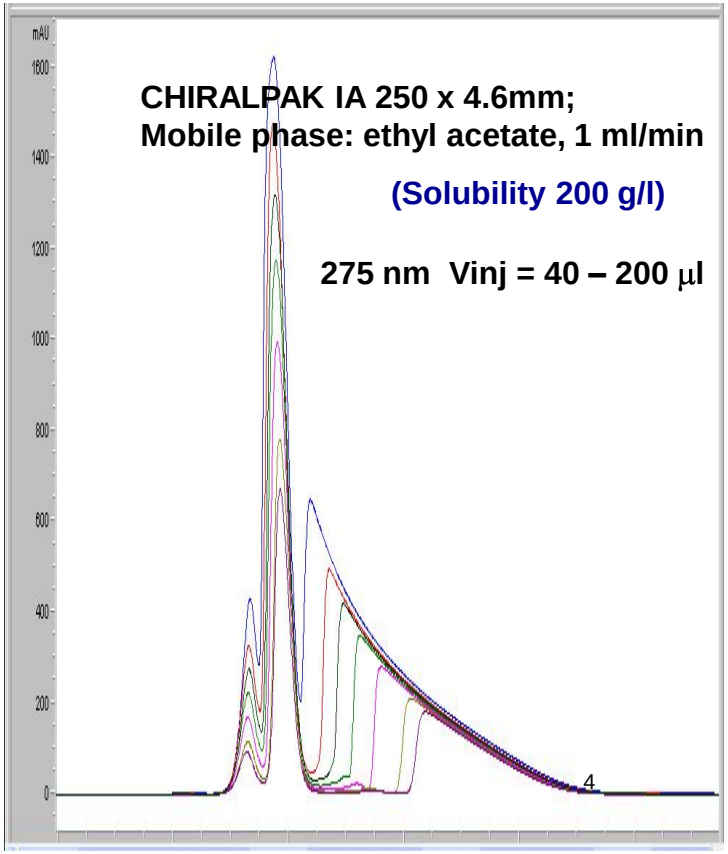
- **Bi-Langmuir isotherm**
- **Overloads rapidly**



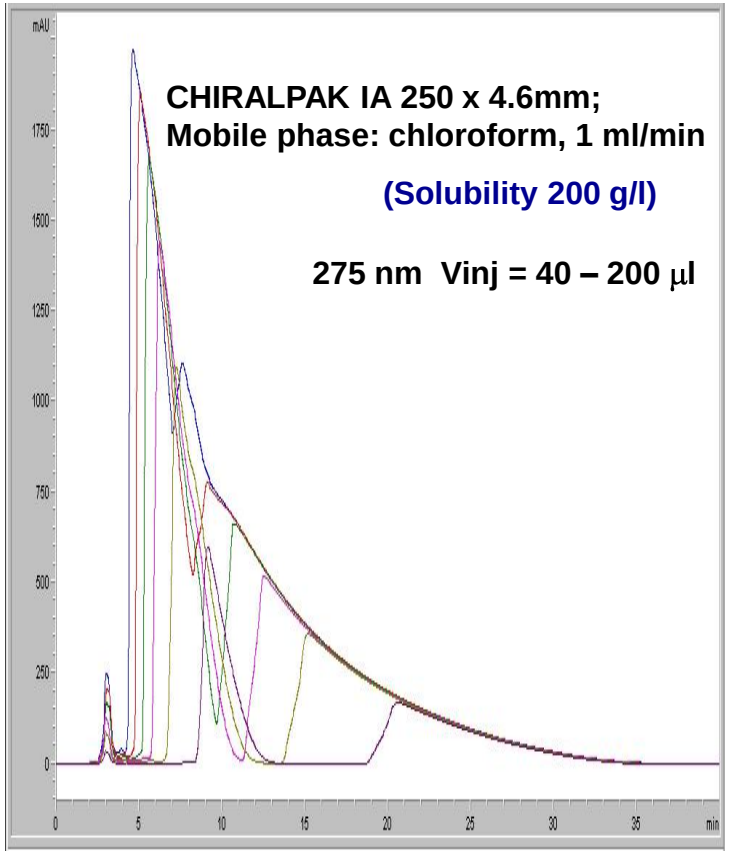


- **Bi-Langmuir/Quadratic Isotherm**
- **Difficult to model**

Loading Studies – EtOAc; $CHCl_3$



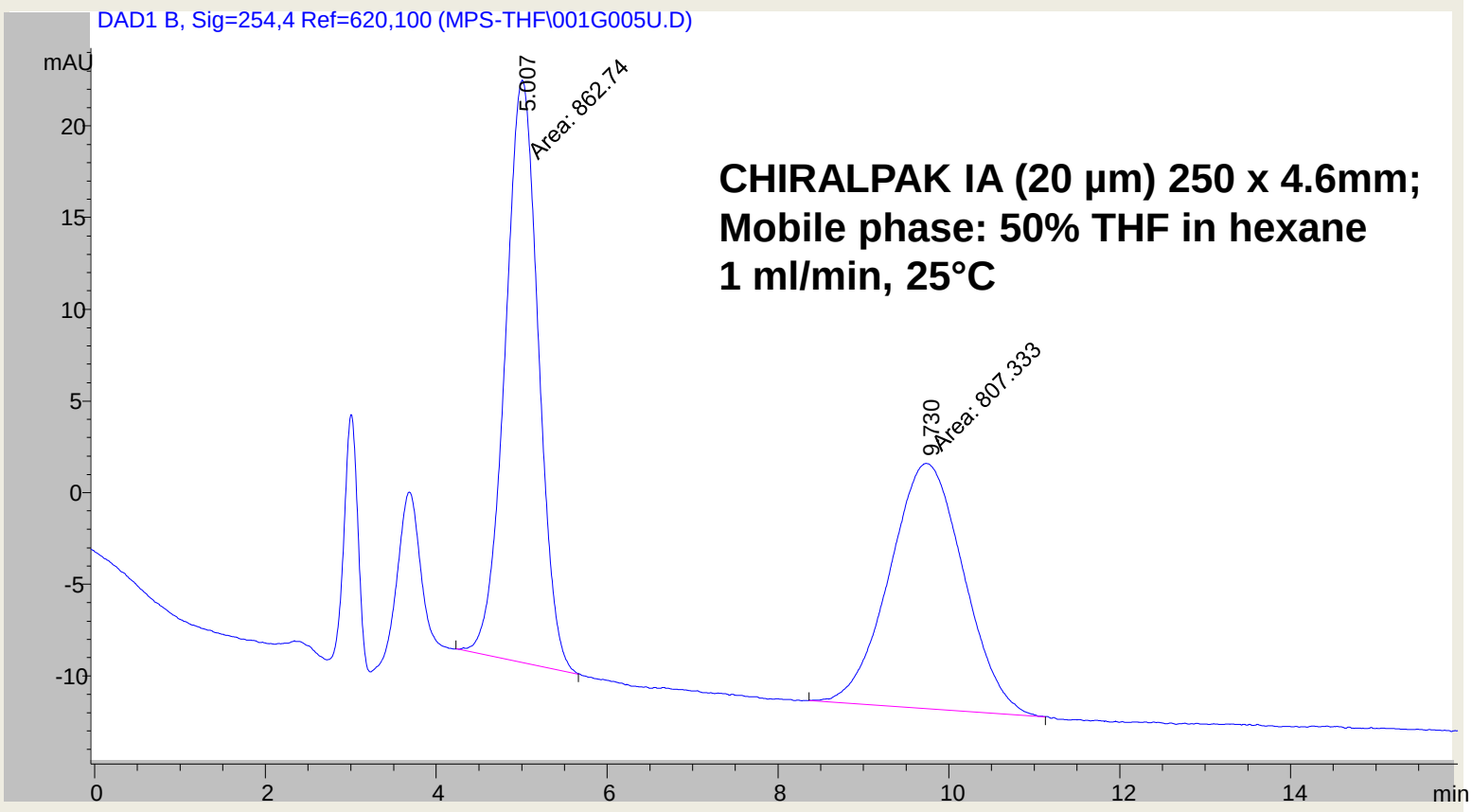
- **Short retention times**
- **Good loading**



- **Long retention times**
- **Overloads rapidly**

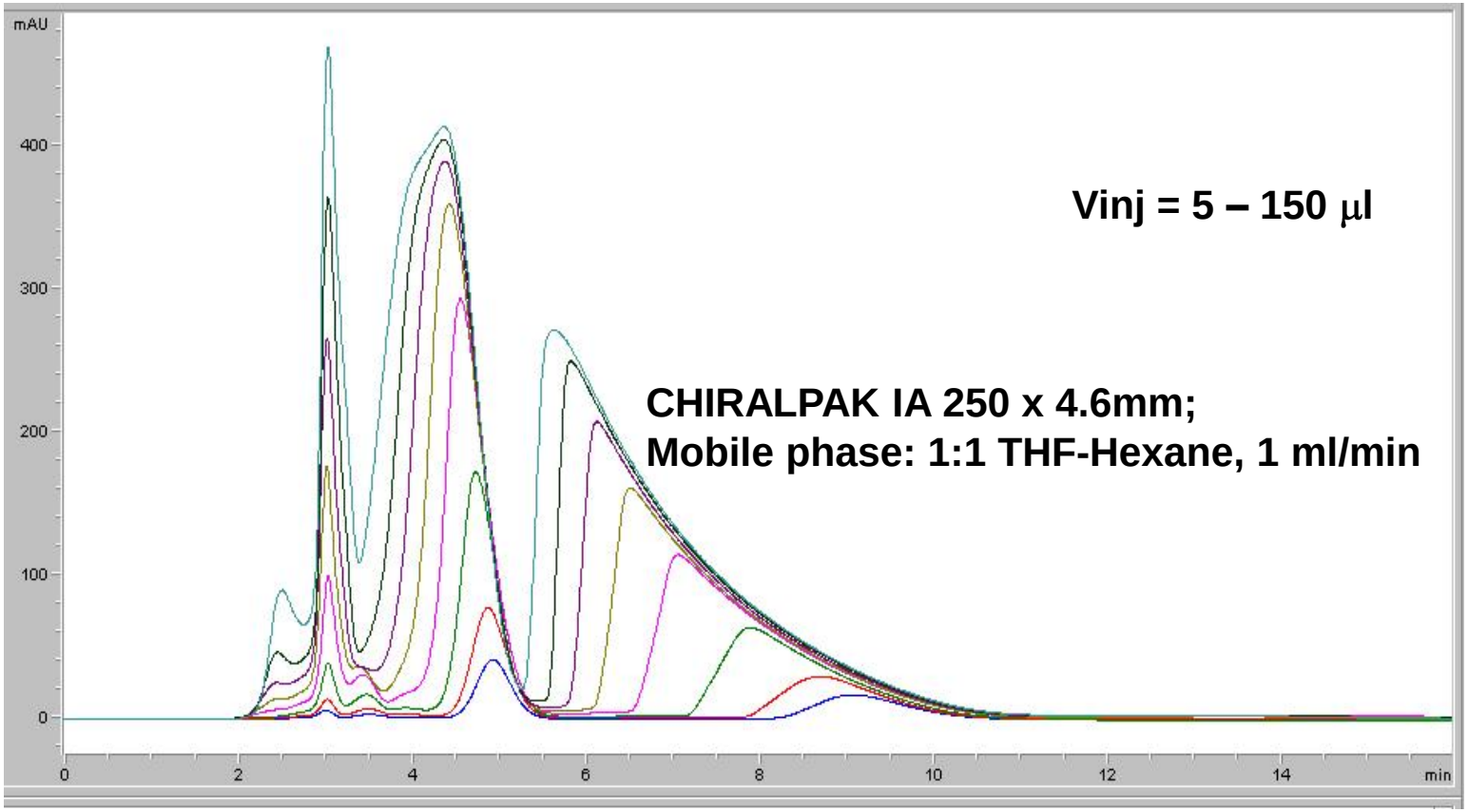
Current Study

Analytical Injection – 1:1 THF-Hexane



Selectivity: 3.35

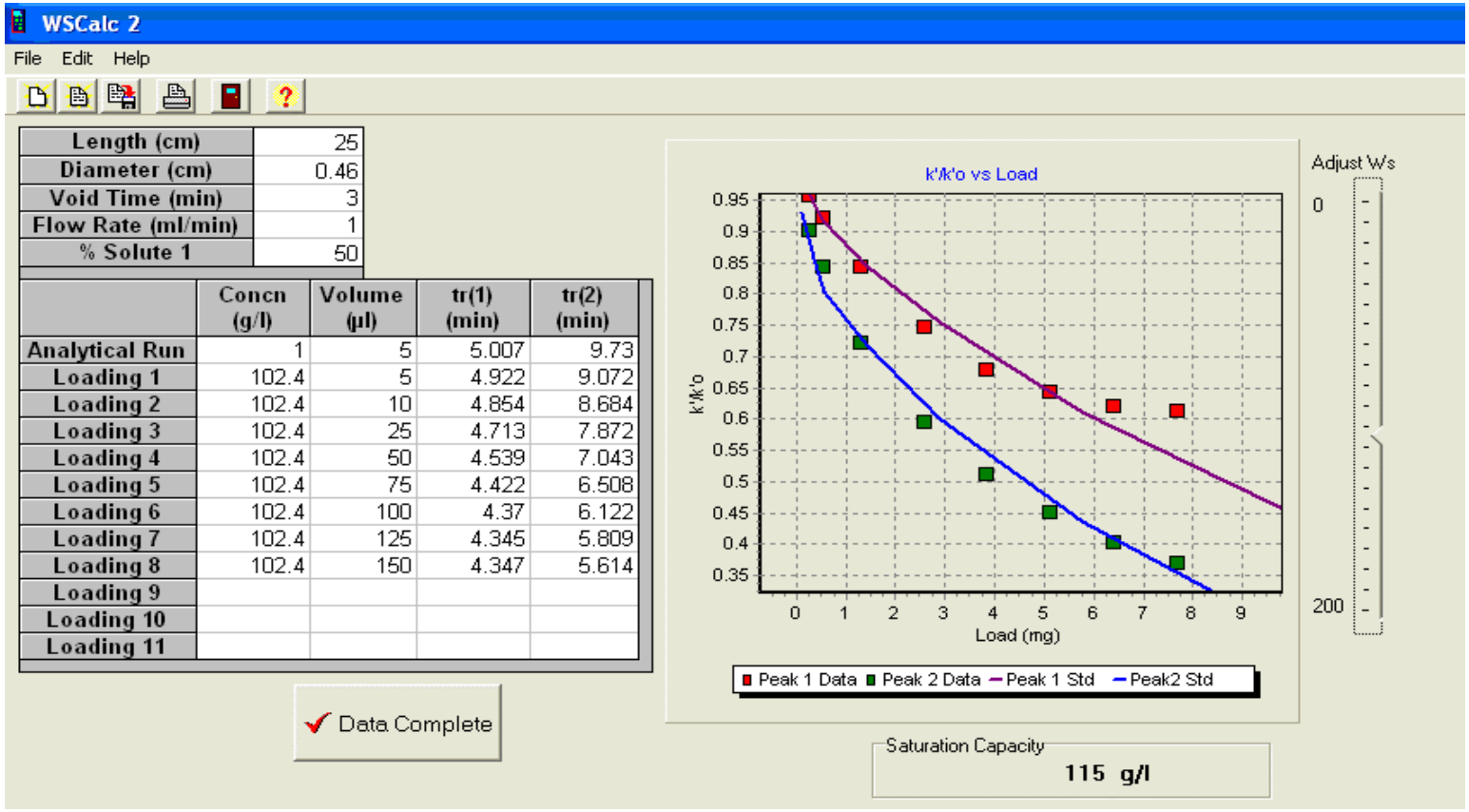
Loading Studies – 1:1 THF / Hexane



Overlaid chromatograms from the loading study.

Loading Data (1:1 THF / Hexane):

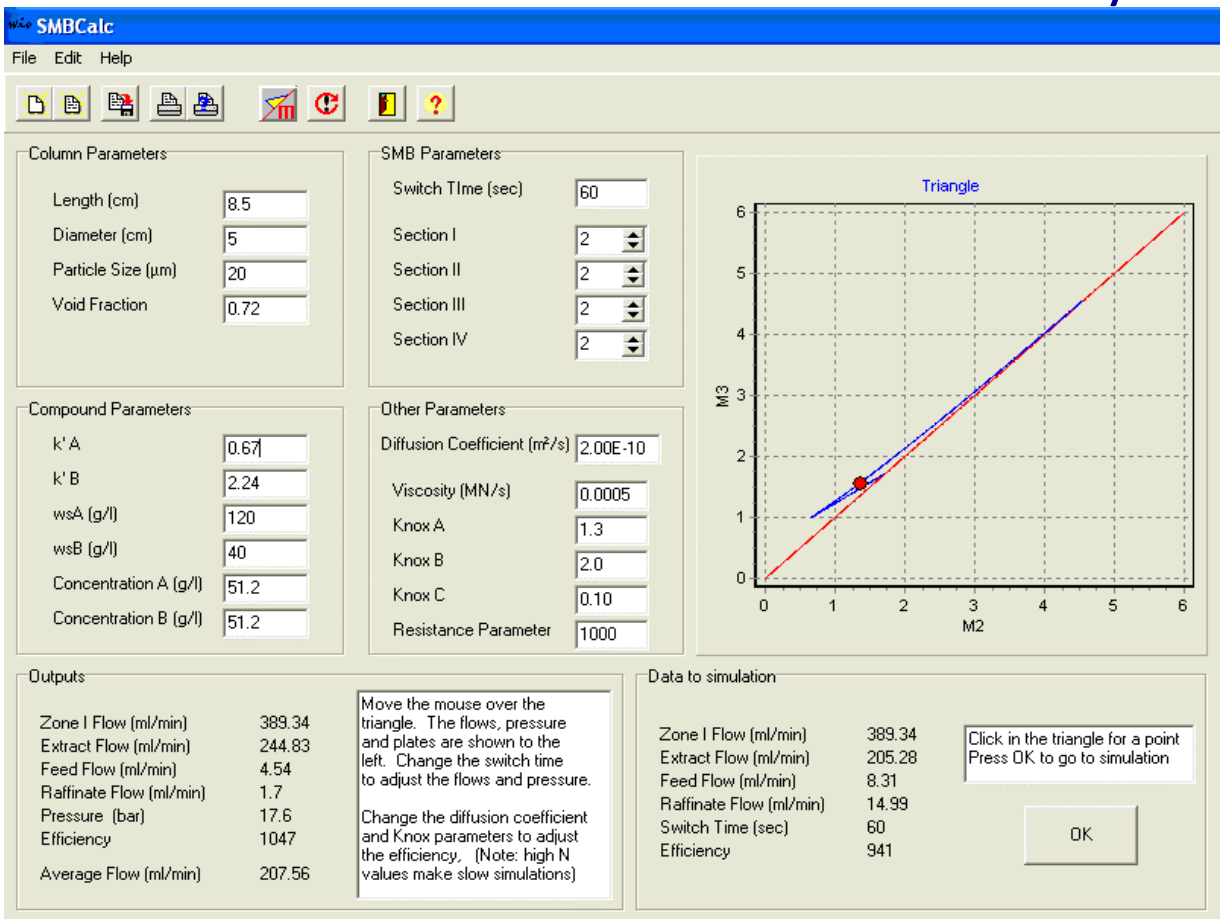
Best Isotherm Fit (Langmuir / 150 -50 IAS)



The isotherm parameters were estimated by adjustment of the saturation capacities to fit simulated retention times to the experimental data.

SMB Parameters:

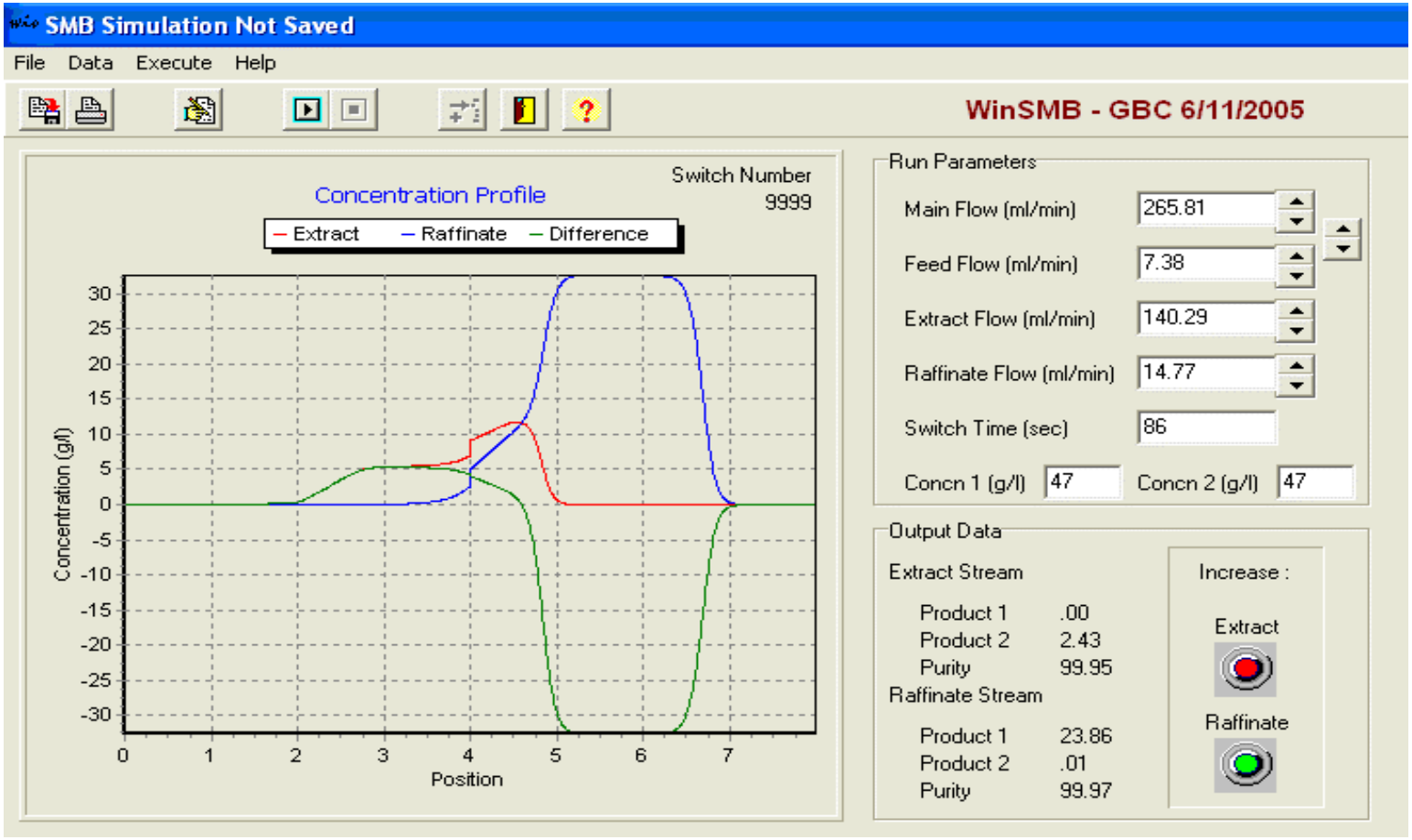
Initial Conditions for a 5 cm System



The software calculates the Morbidelli triangle assuming Langmuir isotherm behavior from input data and uses this to generate SMB conditions for a user-selected point in the operating space. These conditions are used for the computer simulation of the process…

SMB Parameters:

Optimized Conditions for a 5 cm System



Note: Green trace is predicted polarimeter output

Simulation Results:

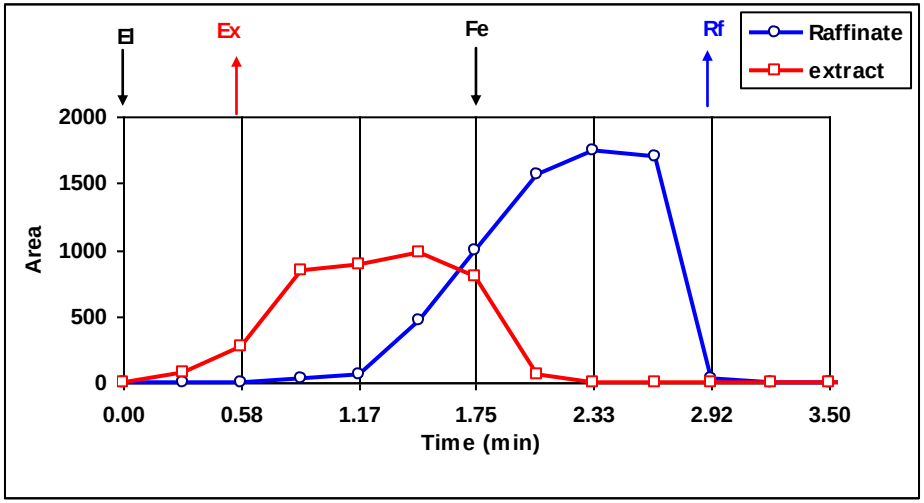
Final Prediction for a mini-SMB System

8 columns (4.6 x 100 mm)	**7.32**	**g CSP**
Feed Flow	**0.11**	**ml/min**
Feed Concentration	**94**	**g/l**
Recycle Flow	**3.91**	**ml/min**
Extract Flow	**2.06**	**ml/min**
Raffinate Flow	**0.22**	**ml/min**
Eluent Flow	**2.17**	**ml/min**
Switch Time	**0.82**	**min**
Zone 1 Flow	**3.91**	**ml/min**
Zone 2 Flow	**1.85**	**ml/min**
Zone 3 Flow	**1.95**	**ml/min**
Zone 4 Flow	**1.74**	**ml/min**
Average Flow Rate	**2.36**	**ml/min**
Extract Purity	**99.03**	**%e.e.**
Extract Concentration	**2.47**	**g/l**
Raffinate Purity	**99.50**	**%e.e.**
Raffinate Concentration	**23.48**	**g/l**
Production Rate	**7.35**	**g/day enantiomer**
Productivity	**1.00**	**kg/kg/day enantiomer**

Experimental Set-Up

- **Columns: Eight 100 x 4.6 mm (each containing 0.915g)**
- **Initial SMB Run (optimized for an operating pressure of 35-40 bar)**
  - **Feed Flow Rate = 0.15 ml/min (Feed at 94 g/l)**
  - **Eluent Flow Rate = 5.50 ml/min (⇔ to Section I Flow)**
  - **Extract Flow Rate = 2.90 ml/min**
  - **Raffinate Flow Rate = 0.31 ml/min**
  - **Switch Time = 35 Seconds**
  - **Recycle stream initially discarded.**
- **Sample Collection after Reaching Steady State**
  - **At outlet ports (Raffinate, Extract and Recycle) during a full cycle.**
  - **At online sampling loop (twice every switch) for a full cycle.**
- **Fraction Analysis: Agilent 1100 System**
  - **Column: CHIRALPAK®IA®, 20 μ, 250 x 4.6 mm**
  - **Mobile phase: 50% THF in Hexane, Flow = 1 mL/min, P = 4 bar, T = 25°C**
  - **Injection Volume = 10 μl, Detection: UV at 270 nm**

Experimental Internal Profile for MPS/IA System



CHIRALPAK IA / 1:1 Hexane : THF

Experimental Optimal SMB Parameters (THF/Hexane)

SMB Parameters

	Experimental	From Simulation
Zone1 (Eluent):	**9.20 ml/min**	**8.87 ml/min**
Feed (@ 94 g/l):	**0.24 ml/min**	**0.25 ml/min**
Extract:	**5.03 ml/min**	**4.68 ml/min**
Raffinate:	**0.75 ml/min**	**0.50 ml/min**
Switch Time:	**0.58 min**	**0.36 min***

**Note: Poor correlation of simulation with experiment*

Other Parameters

Amount of CSP in 6 columns:	**5.49 g**
Column Dimension:	**100 x 4.6 mm**
Number of Columns:	**6**
Operating Pressure:	**32-41 bar**

Productivity ($kg_{enantiomer}$ / kg_{CSP} / Day) = 2.96

Comparison of Separations - SMB

Mobile Phase:	MTBE	ACN/IPA	EtOAc	$CHCl_3$	THF/Hex
Viscosity (mPa.s)	0.27	0.63	0.45	0.57	0.38
Solubility (g/l)	35	70	200	200	102
Selectivity	3.62	6.83	5.17	3.17	3.35
Sat. Capacity (g/l)	***	54/22	250/59	55/45	120/40
Pressure (bar)	34*	30-37**	35-41**	35+	32-41**
SMB Productivity (kg/kg/day)	2.16*	0.98**	4.97**	0.69+	2.96**

** Values from experiment normalized for pressure*

*** Values from experiment*

**** Saturation capacity is a meaningless parameter for the isotherm observed*

+ Values from simulations only

Conclusions

- **Use of immobilized CSPs in Prep chromatography allows:**
  - **Choice of solvent to give high solubility**
  - **Choice of solvent to give best selectivity**
  - **Choice of solvent/CSP combination with best isotherm properties to maximize production**
- ***The best choice of solvent is not always obvious!***