

Twin-column counter-current chromatography for purification of biologics

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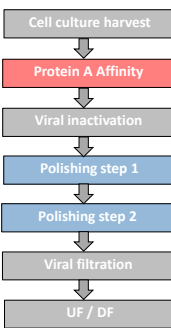
Summary

> Twin-column chromatography processes have been used for capture and polishing applications, outperforming single column batch chromatography in terms of productivity, yield, buffer consumption

> Suitable for continuous downstream processing

> Dynamic UV-based process control ("AutomAb") included

> Operation with only two columns offers the full benefits of multicolumn chromatography while minimizing equipment complexity



CaptureSMB instead of batch capture process:

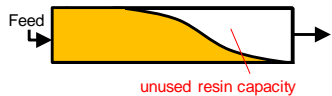
Typical result:
40-60% protein A savings
40-60% buffer savings
40-60% increase of product concentration
2-3x productivity increase

MCSGP instead of batch polishing process:

Typical result:
50% yield increases
50% buffer savings
40-60% increase of product concentration
2-3x productivity (peptide up to 10x) increase

CaptureSMB process principle

> Single column Batch Capture: A large fraction of the affinity resin remains unused



→ Used capacity typically 40-60% of static binding capacity (SBC).

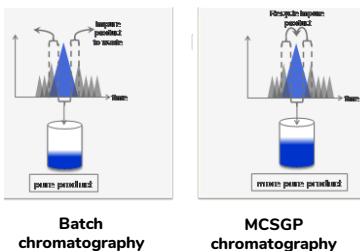
> Sequential loading processes: By splitting the bed into two columns, the capacity utilization is maximized.



→ Used capacity typically > 90% of SBC
→ Faster loading flow rates can be used

MCSGP process principle

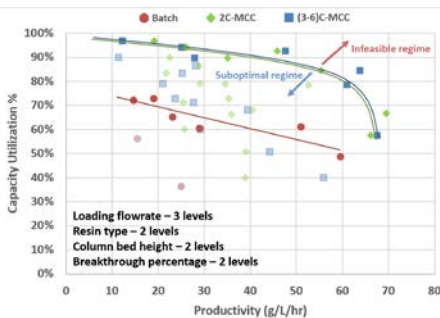
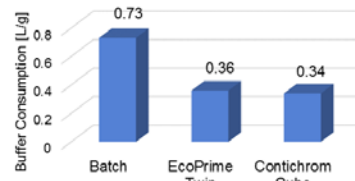
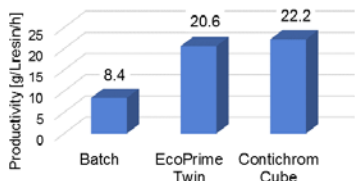
MCSGP (Multicolumn Countercurrent Solvent Gradient Purification) is a scalable twin-column chromatographic purification process based on the internal recycling of partially pure side-fractions to obtain high yield / high purity simultaneously.



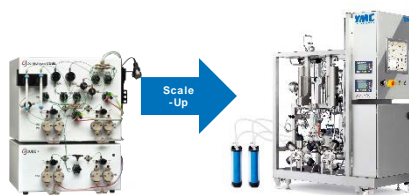
CaptureSMB scale-up

Twin-column scale-up study with BMS at 5 g/L titer showed

- 2.5x productivity improvement over single column capture, comparable product quality
- Excellent scalability from Contichrom to EcoPrime Twin scale-up system



Data presented by BMS at ACS Meeting, New Orleans, March 2018



ChromaCon
A new dimension in purification

YMC
PROCESS TECHNOLOGIES

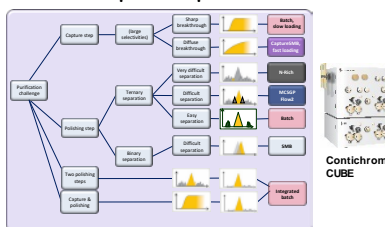
Contichrom® CUBE

EcoPrime Twin GMP

System/Model	Columns (Ø Range)	Feed per day (L/day)	Product recovery (%)
SP Twin 100 (Ø 100)	2,5	50	73
SP Twin 200 (Ø 150)	5	100	80
SP Twin 300 (Ø 200)	8	150	85
SP Twin 400 (Ø 250)	12	200	88
SP Twin 500 (Ø 300)	15	250	90

- Operates with 2 (twin) columns (0.5 – 2.0 cm I.D.)
- Flow rate 0.1-36 mL/min / 0.1-100 mL/min
- Pressure rating 50 bar, 725 psi / 100 bar, 1500 psi
- Compatible with all stationary phases, aqueous buffers and common RP solvents, linear solvent gradient capabilities
- ChromIQ® operating software
- Supports dynamic process control (AutomAb, Mcontrol)
- Active buffer management
- Wizards for easy transition from batch to continuous

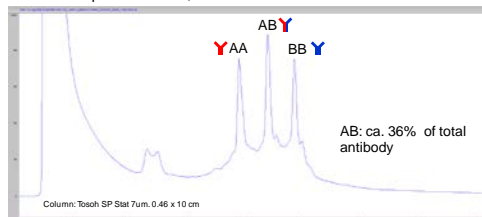
Overview of process capabilities



MCSGP bispecific antibody purification

Cell culture supernatant contains:

- > target antibody AB
- > parental antibodies AA, BB
- > aggregates
- > impurities: HCP, DNA

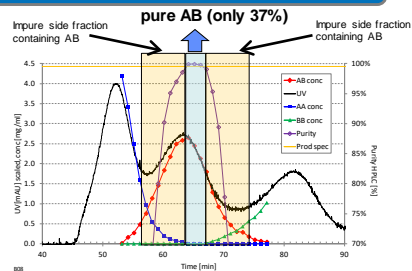


Analytical chromatogram (CIEX) of clarified harvest (cCCS)

Protein A capture

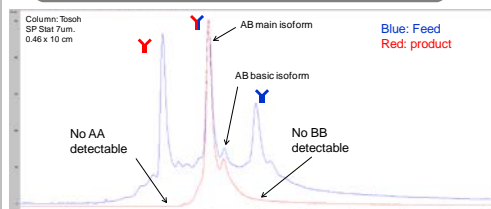
Presence of parental monoclonal antibodies complicates isolation of product with high yield and purity

MCSGP chromatography



Prep chromatogram (batch-CIEX) with pure product fraction and side fractions

Product quality



Analytical chromatogram (CIEX) of product from of 1st polishing step (MCSGP)

Process Performance

The purity specifications were met:

- Less than 0.5% main isoform content of AA, BB in product
- Less than 30 ng/mg HCP, less than 1.0% aggregates

Process	Yield MCSGP [%]	Yield Batch [%]
Capture	97%	97%
CIEX	87%	37%
AIEX	93%	34%
Total	78%	34%

MCSGP:
Increase of yield by from 37 to 87%,
Reduction of buffer consumption by 25%

Acknowledgements:

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