

Viral Clearance Strategy for POROS Hydrophobic Interaction Chromatography

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Bioprocessing

INTRODUCTION

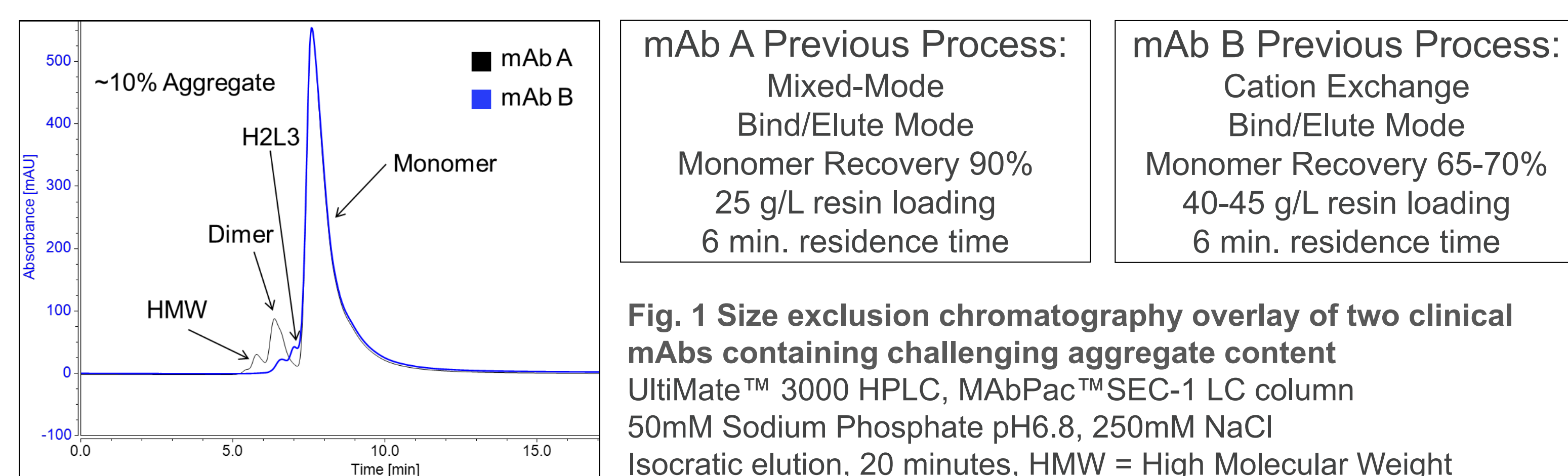
Viral contamination is one of the major concerns in the production of biologics. Next to specific viral reduction steps in the process, every chromatography step in the downstream process may contribute to virus reduction. During process development and at critical clinical stages, it is important to obtain information on the viral clearance properties of each resin used in the purification process. In this study we examined viral clearance on Thermo Scientific™ POROS™ Hydrophobic Interaction Chromatography resins.

STUDY DESIGN

- Virus spiking in CHO cell culture with Mouse Minute Virus (MMV – non-enveloped virus) and Xenotropic Murine Leukemia-related virus (XMuLV – enveloped virus)
- Purification (polish) performed with all three POROS HIC resins on either mAbA or mAbB:
 - POROS Ethyl
 - POROS Benzyl
 - POROS Benzyl Ultra

CASE STUDY – VIRAL CLEARANCE IN HIC MAB POLISHING

POROS HIC POLISHING STEP DEVELOPMENT FOR TWO CLINICAL mAbs:
WHAT VIRAL CLEARANCE CAN BE EXPECTED?



PROCESS OPTIMIZATION mAb A – BIND/ELUTE AND FLOW-THROUGH

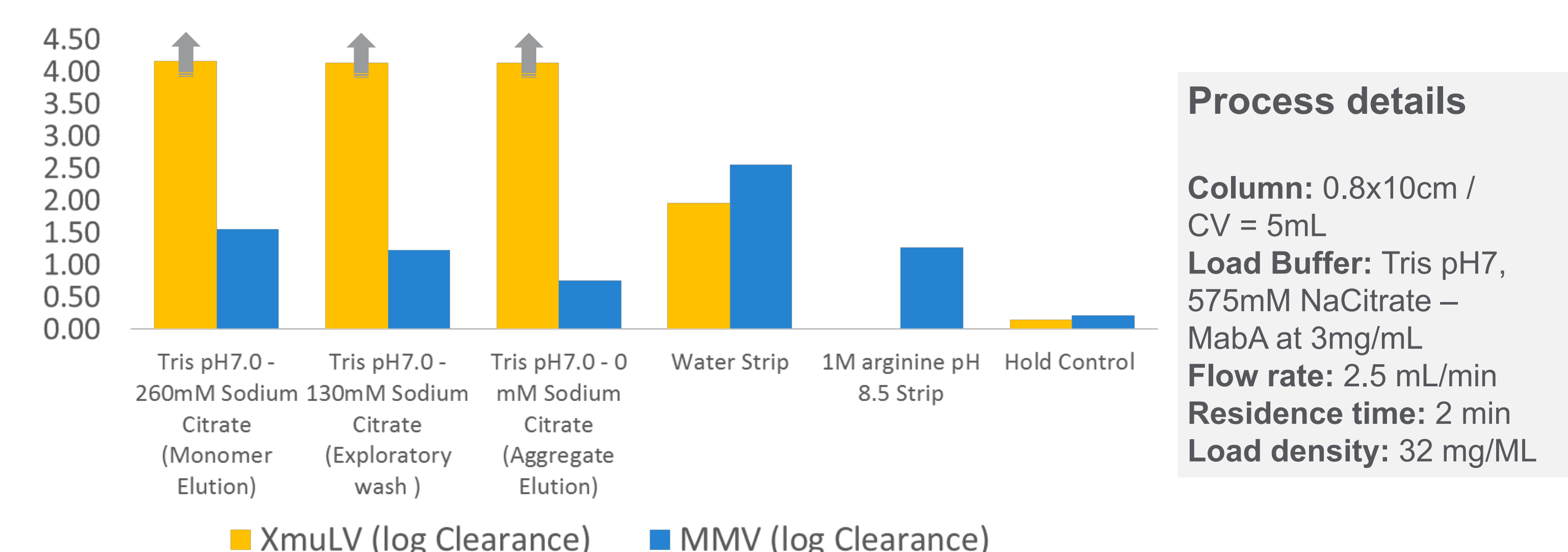
POROS HIC HIGH THROUGHPUT SCREENING FOR mAb A

Resin type	POROS Ethyl			POROS Benzyl			POROS Benzyl Ultra					
	Sodium Chloride	Sodium Acetate	Ammonium Sulfate	Sodium Chloride	Sodium Acetate	Ammonium Sulfate	Sodium Chloride	Sodium Acetate	Ammonium Sulfate			
Partition Selectivity				2.43	1.84							
				3.12	2.74	2.18						
			2.48	4.19	2.59	2.41	2.35	2.38	2.07			
					2.84	2.52	2.70	2.63	2.20	2.27	2.03	
					2.43		2.66	3.53	2.45	3.12	2.02	2.19
							2.54	3.79	2.39	3.73	2.55	2.68
								3.12	3.46	4.34	3.76	
								5.36	4.23	7.17	8.16	

POROS Benzyl Chosen for Bind-Elute optimization, Sodium Citrate Chosen as Salt Type

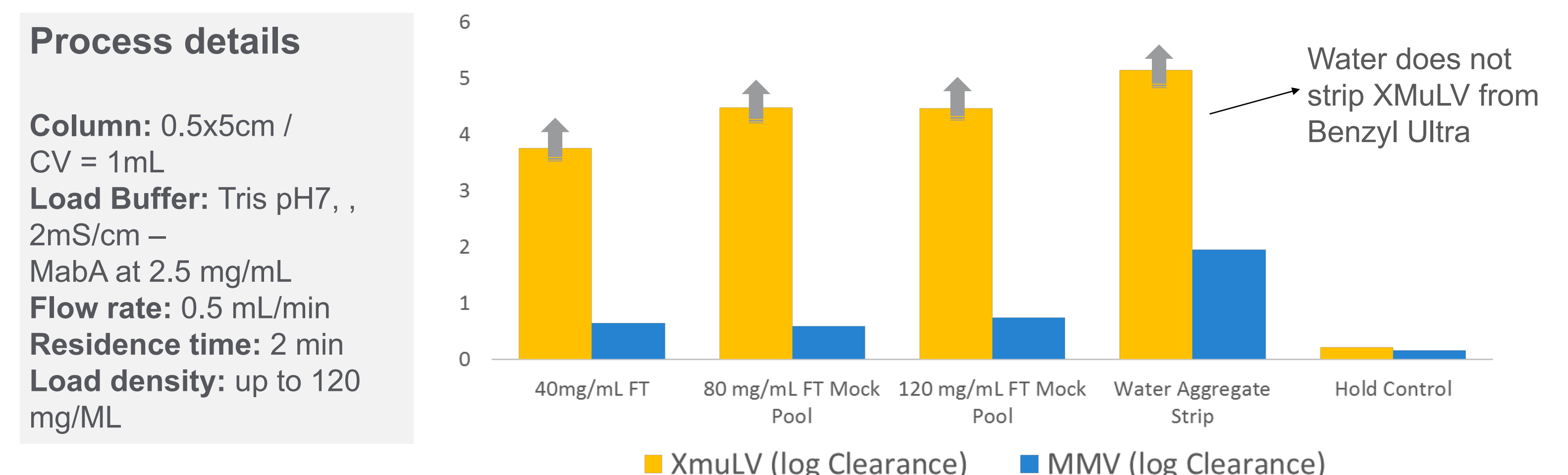
POROS Benzyl Ultra Chosen for Flow-through optimization under no salt

POROS BENZYL BIND/ELUTE - VIRAL CLEARANCE FOR mAb A



- ✓ Complete XMuLV (>4LOG) clearance in mAb A pool
- ✓ Partial MMV clearance in mAb A pool
- ✓ Water partially and Arginine fully strips XMuLV from POROS Benzyl

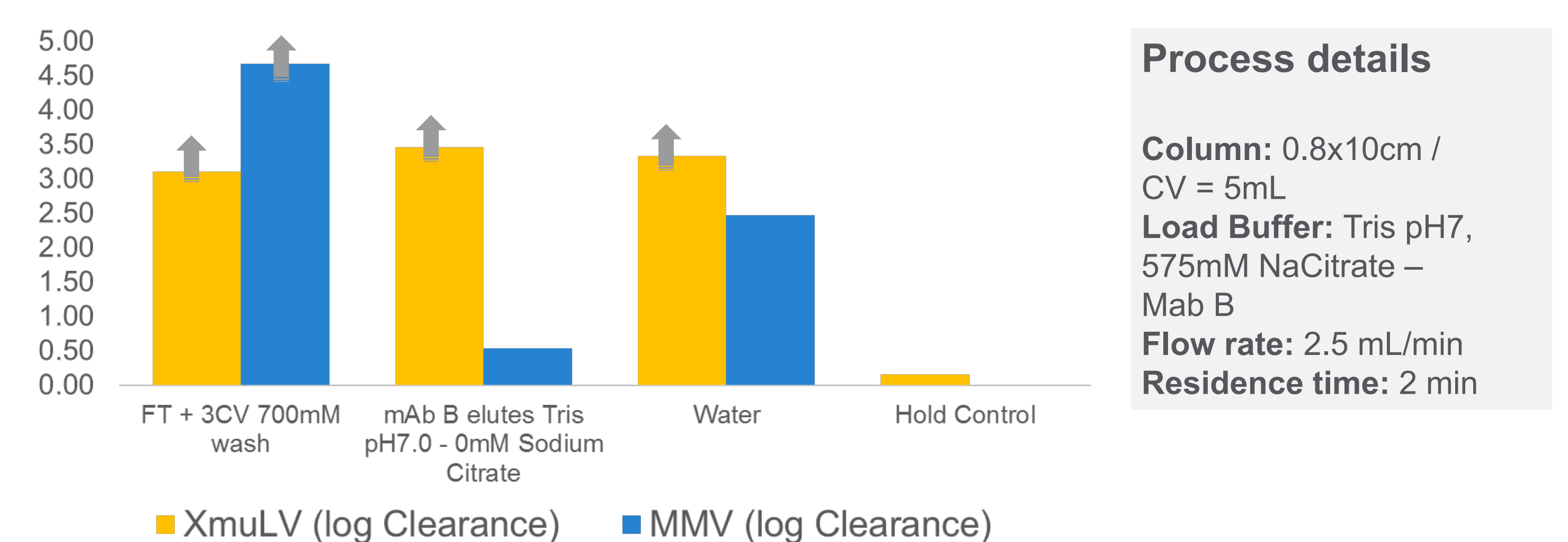
POROS BENZYL ULTRA FLOW-THROUGH VIRAL CLEARANCE FOR mAb A



- ✓ Complete XMuLV (>4LOGS) clearance in mAb A Flow-Through
- ✓ Minimal MMV (~0.5 LOG) clearance in mAb A Flow-Through

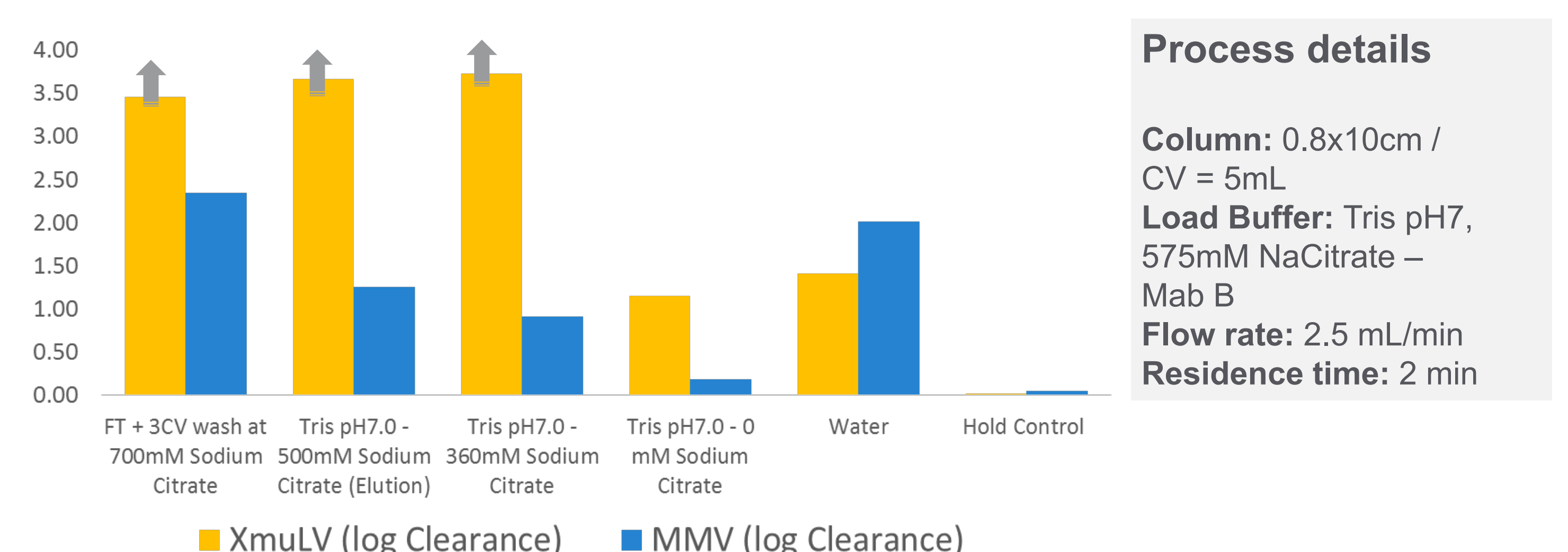
PROCESS OPTIMIZATION mAb B – CHALLENGING H2L3 AGGREGATE

POROS BENZYL ULTRA BIND/ELUTE - VIRAL CLEARANCE FOR mAb B



- ✓ Complete XMuLV (>3 LOGS) clearance in mAb B pool
- ✓ Minimal MMV (~0.5 LOG) clearance in mAb B POOL
- ✓ Buffer with no salt elutes MMV

POROS ETHYL BIND/ELUTE VIRAL CLEARANCE FOR mAb B



- ✓ Complete XMuLV (>4 LOGS) clearance in mAb B pool
- ✓ Partial MMV (1.3 LOG) clearance in mAb B pool
- ✓ Buffer with no salt elutes MMV and XMuLV

CONCLUSIONS

- Complete XMuLV viral clearance on all POROS HIC resins in both bind/elute (Benzyl & Ethyl) as in flow-through mode (Benzyl Ultra)
- Partial MMV viral clearance due to lower hydrophobicity of the virus
- Despite moderate MMV log reduction, these data indicate MMV log reduction could be improved with further optimization

TRADEMARKS/LICENSING

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