

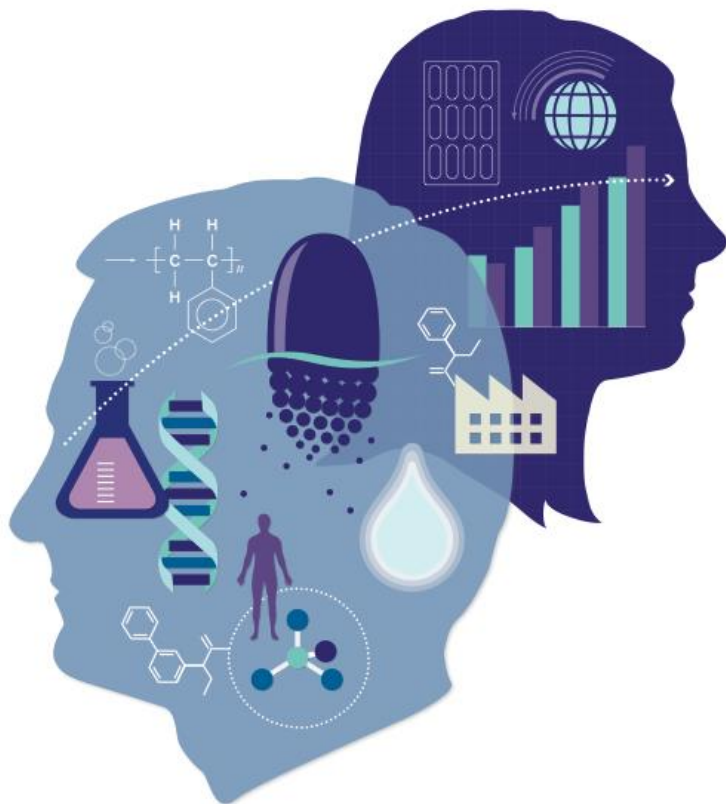
About OMICS Group

OMICS Group is an amalgamation of [Open Access Publications](#) and worldwide international science conferences and events. Established in the year 2007 with the sole aim of making the information on Sciences and technology 'Open Access', OMICS Group publishes 700+ online open access [scholarly journals](#) in all aspects of Science, Engineering, Management and Technology journals. OMICS Group has been instrumental in taking the knowledge on Science & technology to the doorsteps of ordinary men and women. Research Scholars, Students, Libraries, Educational Institutions, Research centers and the industry are main stakeholders that benefitted greatly from this knowledge dissemination. OMICS Group also organizes 1000+ [International conferences](#) annually across the globe, where knowledge transfer takes place through debates, round table discussions, poster presentations, workshops, symposia and exhibitions.

OMICS International Conferences

OMICS International is a pioneer and leading science event organizer, which publishes around 700+ open access journals and conducts over 500 Medical, Clinical, Engineering, Life Sciences, Pharma scientific conferences all over the globe annually with the support of more than 1000 scientific associations and 30,000 editorial board members and 3.5 million followers to its credit.

OMICS Group has organized 1000+ conferences, workshops and national symposiums across the major cities including San Francisco, Las Vegas, San Antonio, Omaha, Orlando, Raleigh, Santa Clara, Chicago, Philadelphia, Baltimore, United Kingdom, Valencia, Dubai, Beijing, Hyderabad, Bengaluru and Mumbai.



Compatibility Assessment of a Model Monoclonal Antibody Formulation in Glass and in Blow-Fill-Seal (BFS) Plastic Vial Delivery Formats

Dipesh Shah, Ph.D.
Senior Development Engineer
Advanced Delivery Technologies,
Woodstock, Illinois

©2014 Catalent Pharma Solutions. All rights reserved



Presentation Outline

- 1. Advanced Aseptic Delivery Technology: Blow-Fill-Seal*
- 2. General Requirements for Container Qualification and examples of Protein-Container Interactions*
- 3. Case – Study: Compatibility assessment of a model monoclonal antibody formulation with Advasept™ and Glass vial*



1: Advanced Aseptic Processing: Blow Fill Seal



DEVELOPMENT



DELIVERY



SUPPLY

more products. better treatments. reliably supplied.™

Advanced Delivery Technologies for Parenterals to Mitigate Risk Associated with Aseptic Processing

Advanced Aseptic Filling Technology



Why Glass Free?

Quality/Sterility Assurance

Reduce risk factors for sterility challenges through automation

Minimizing Particulates

Elimination of glass particles and delamination with significant reduction in foreign particulates

Safety/Reduced Product Loss

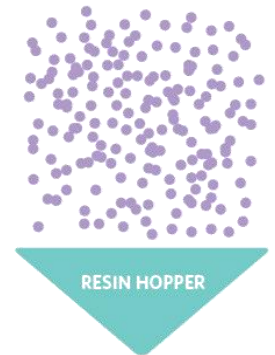
Plastic construction reduces risk of breakage and simplifies opening

Stability/Compatibility

Medical grade polypropylene resin for excellent chemical and physical properties

Minimize Risk: Simplify the Process, Reduce Variables

In just **15 seconds**, the container is formed, filled and sealed in ISO 5 aseptic conditions



TIP

SCREEN

EXTRUDER

PARISON HEAD

MIXING ZONE

METERING ZONE

TRANSITION ZONE

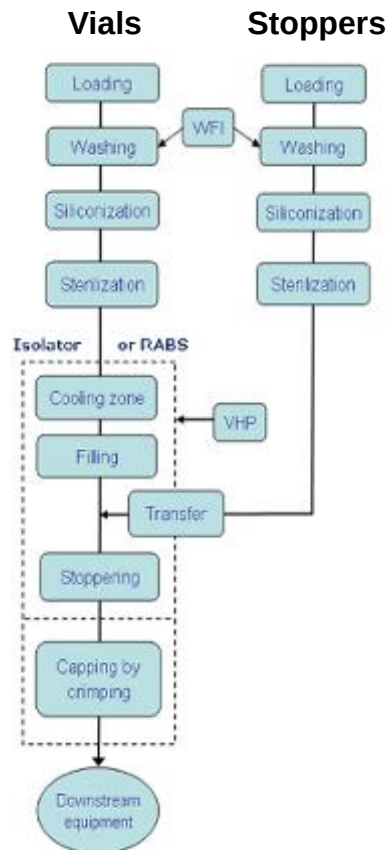
FEED ZONE



Minimizing Variables and the Footprint in Aseptic Manufacturing

Utilizing the automated aseptic design of BFS, the technology eliminates traditional manufacturing steps, reduces the required controlled space and decreases the risk of contamination associated with traditional aseptic practices

Traditional Glass Vial Filling



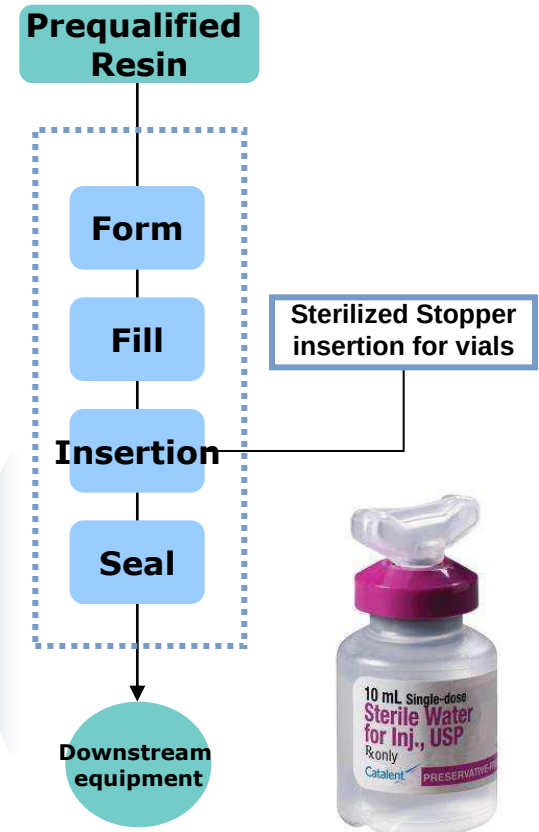
Glass Vial Filling

- Large controlled space
- Glass management and process control

Blow Fill Seal

- Class A space 10 sq. ft
- Sterile container is formed at the time of fill

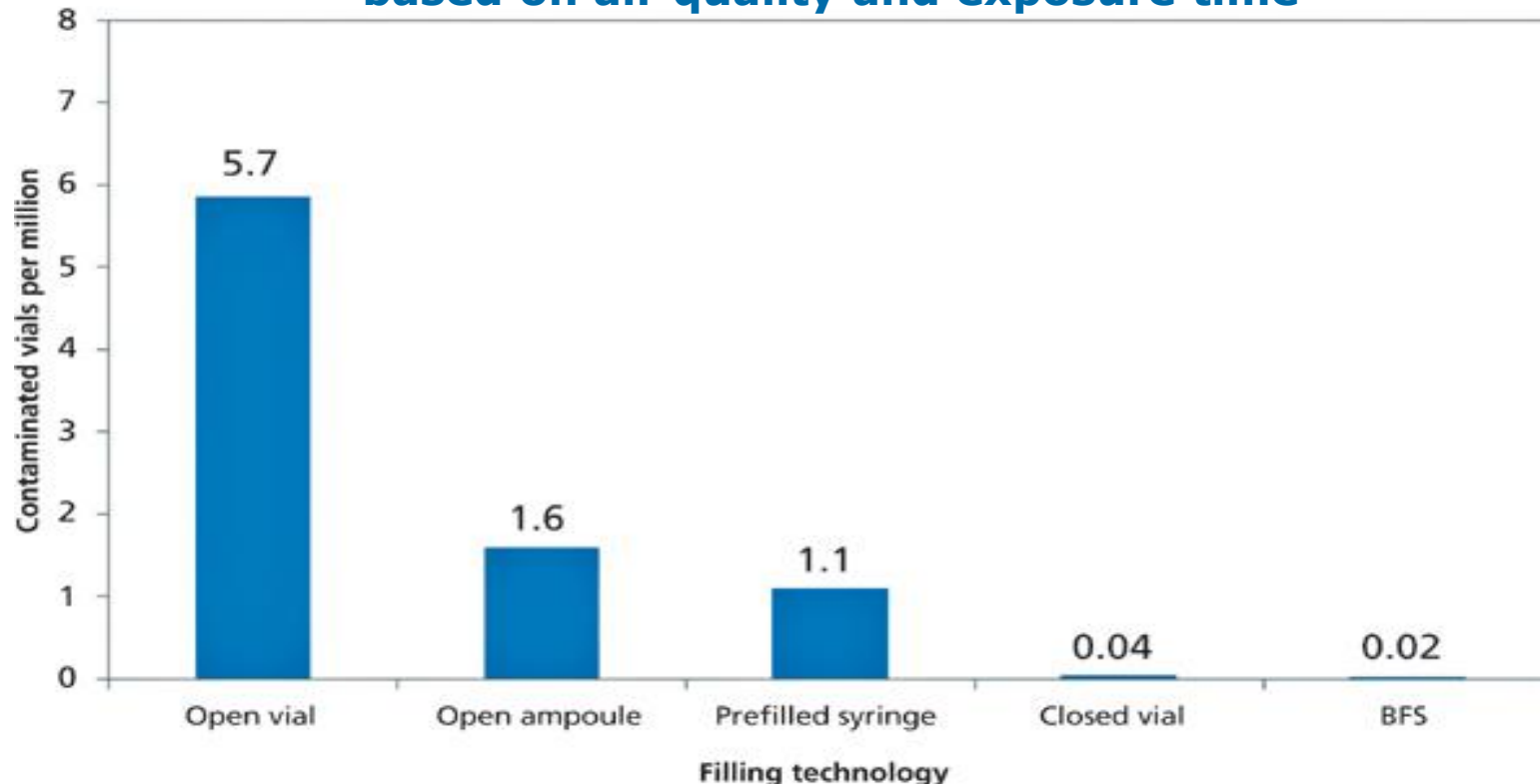
Blow Fill Seal



Leveraging Advanced Aseptic Process Blow Fill Seal Technology

The BFS process minimizes the risk of contamination by reducing particles, process steps & human interaction

Potential risk of contamination by filling technology based on air quality and exposure time



Reference: Verjans, B. Reed, C. (2012). "Assessing Filling Technologies for Contamination Risk." *Biopharm International*. 25(3), pp. 46-58.



2: General Requirements for Container Qualification and Examples of Protein-Container Interactions



DEVELOPMENT



DELIVERY



SUPPLY

more products. better treatments. reliably supplied.™

General Requirements for Container Qualification

Safety –

- Are we maintaining sterility?
- What are we leaching?

Efficacy –

- Are we adequately protecting the dose?
- Is there binding -- adsorption/absorption?
- Are we delivering the dose?

Container Interactions with Protein Therapeutics

Discussion Topics

- 1. What are the product development concerns for container interactions with protein products?*
- 2. What testing is needed to understand container interactions with protein therapeutics?*

Liquid Formulation Containers (Injectables)– Potential Materials

<i>Enhanced System</i>	<i>Container Materials</i>	<i>Closure Materials</i>
<i>Vials or Ampoules (SVP)</i>	<i>Glass/Advasept™</i>	<i>Vials with Butyl rubber/Teflon coated butyl rubber</i>
<i>Prefilled syringes with plungers</i>	<i>Glass or Plastic Staked needle or luer lock (plastic or rubber)</i>	<i>Butyl rubber Teflon coated butyl rubber</i>
<i>IV Infusion (LVP)</i>	<i>Glass/Flexible Bags with spike ports/Advasept™</i>	<i>Polyethylene</i>

General Concerns for Protein-Container Interactions

Container Concerns	Product concerns
<ul style="list-style-type: none">• <i>Protein adsorption</i>	<ul style="list-style-type: none">• <i>Protein loss</i>
<ul style="list-style-type: none">• <i>Headspace</i>	<ul style="list-style-type: none">• <i>Agitation causes protein aggregation (Advasept™ Advantage)</i>
<ul style="list-style-type: none">• <i>Organic leachables (eg. Silicone, DEHP)</i>	<ul style="list-style-type: none">• <i>Toxicity of leachable</i>• <i>Protein aggregation/Immunogenicity</i>
<ul style="list-style-type: none">• <i>*Trace metals (eg. Iron, Tungsten, Aluminum)</i>	<ul style="list-style-type: none">• <i>Eg. Iron – Iron Leachable catalyzed protein degradation reactions</i>• <i>Eg. Tungsten – Protein Oxidation (Advasept™ Advantage)</i>• <i>Eg. Aluminum – Aluminum Phosphate resulted in visible particles (Advasept™ Advantage)</i>

[*www.pqri.org/workshops/podp11/pdfs/markovic.pdf](http://www.pqri.org/workshops/podp11/pdfs/markovic.pdf), “Regulatory perspective safety qualification of extractable and leachable,”

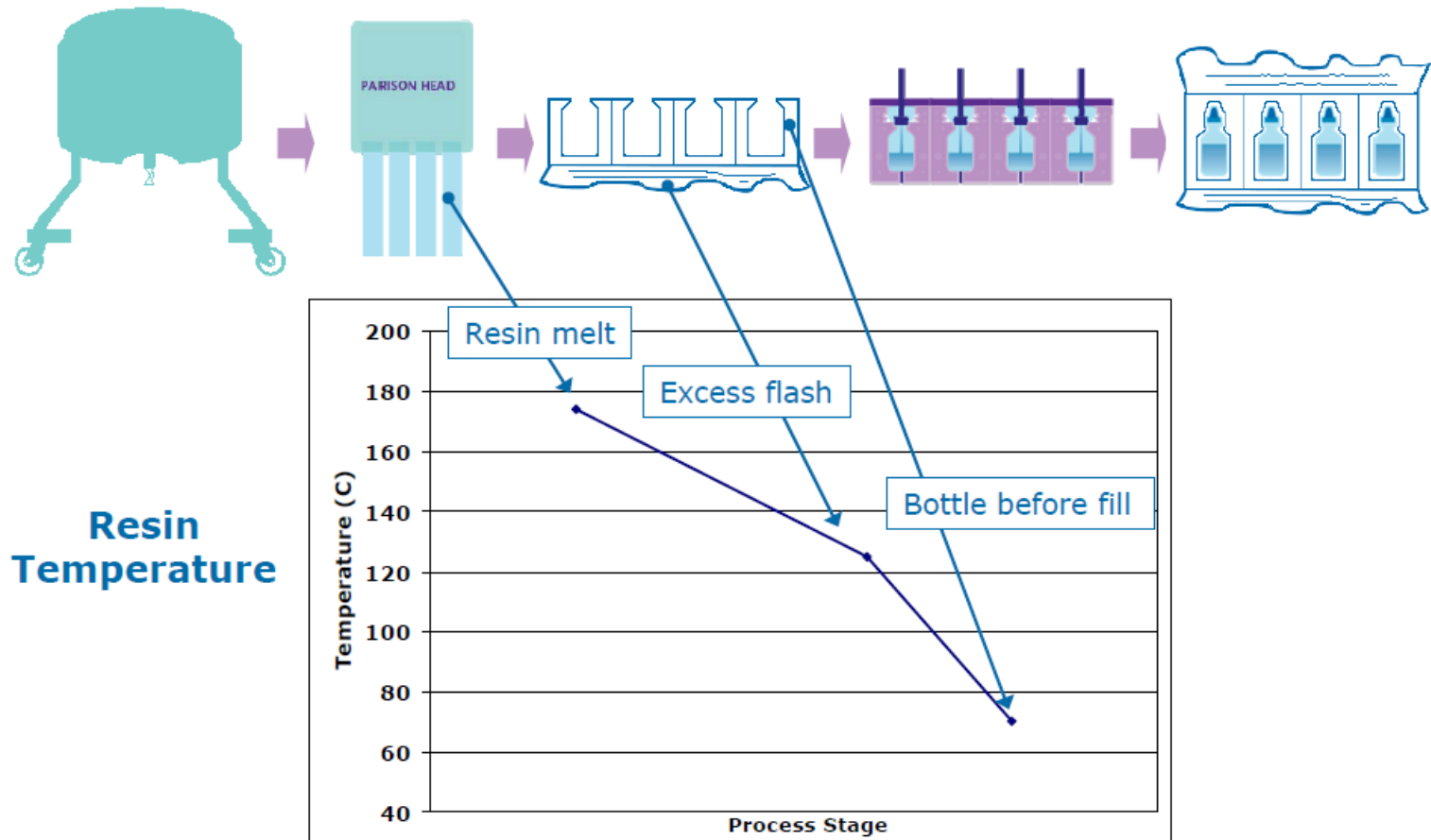
Feb. 22-23, 2011, Ingrid Markovic, Ph.D.

Challenges to fill Biologics with Blow-Fill- Seal (BFS)

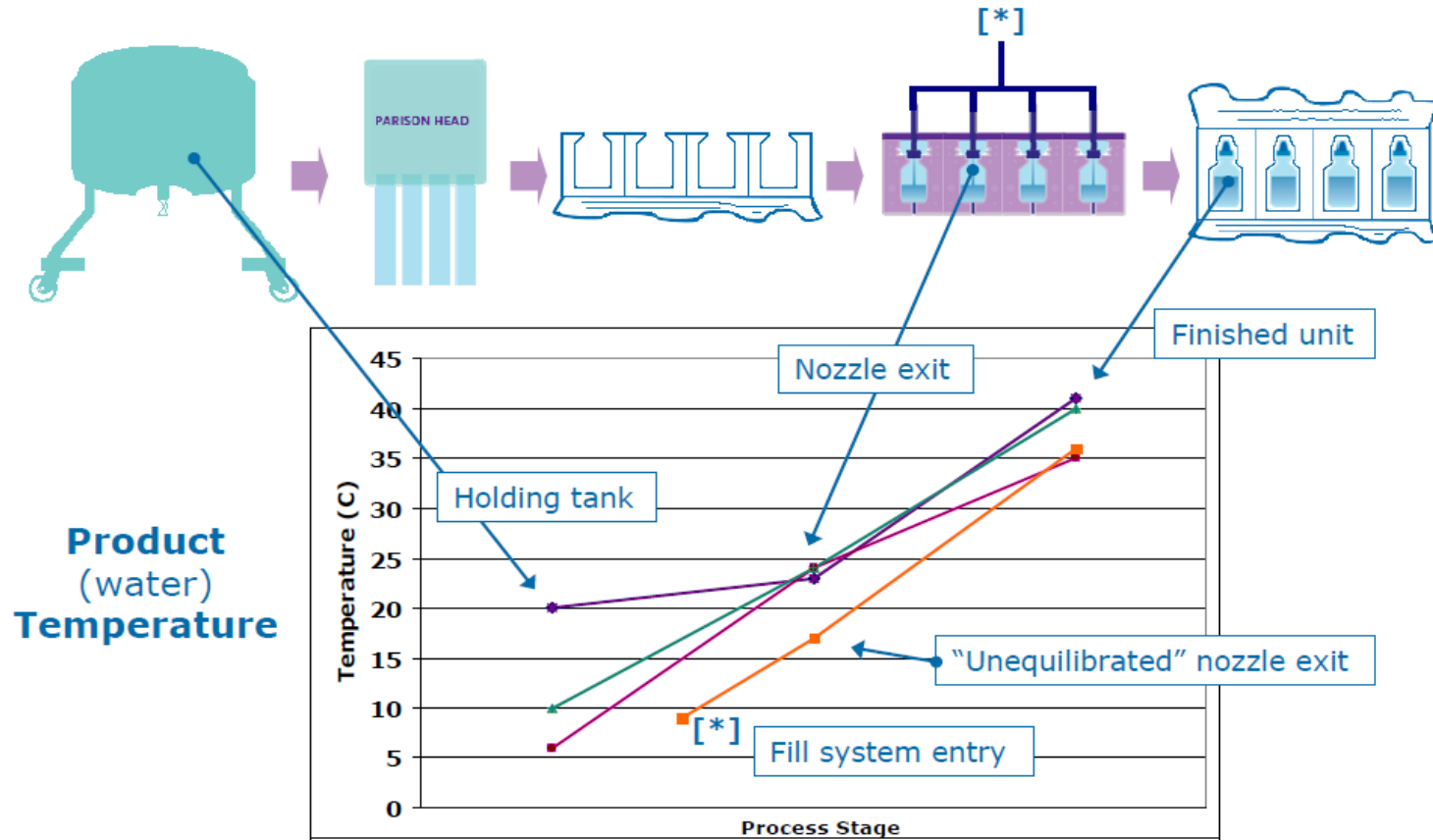
Challenges:

1. Impact of elevated temperature of the molded plastic during filling on the stability of the biologic formulation
2. Impact of gas permeation from semi-permeable BFS plastic container on degradation of biologics (for eg. Oxidation)
3. Impact of potential leachables from BFS processed plastic container system on biologic stability and safety of the formulation.

Temperature Profile of Resin and Container

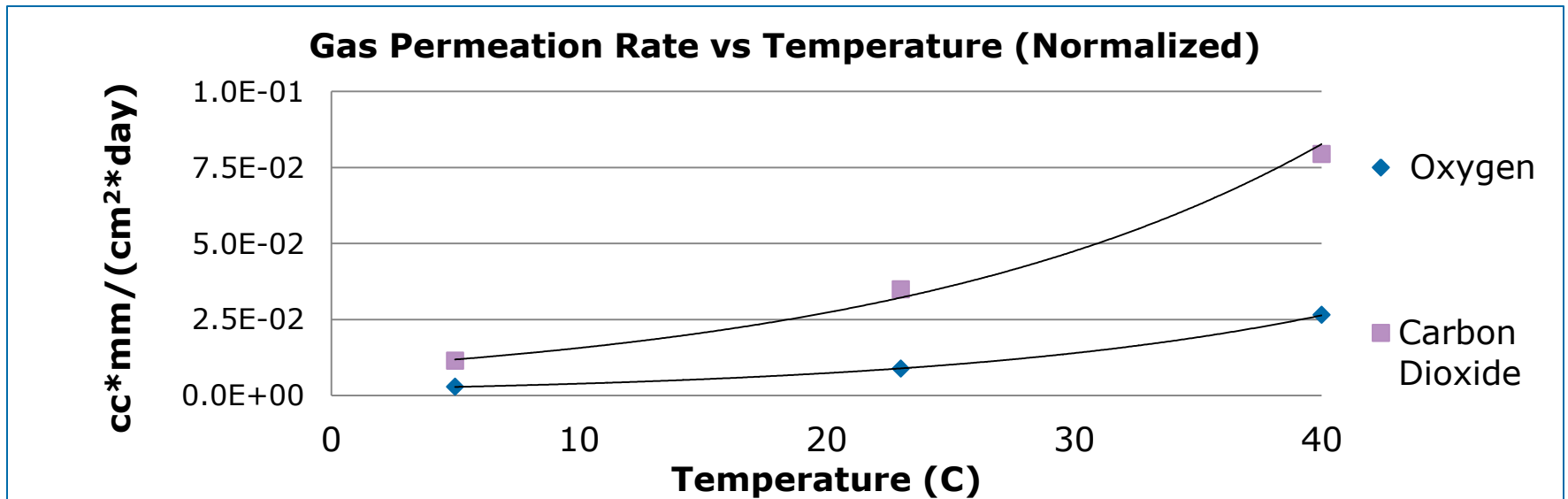


Temperature Profile of solution



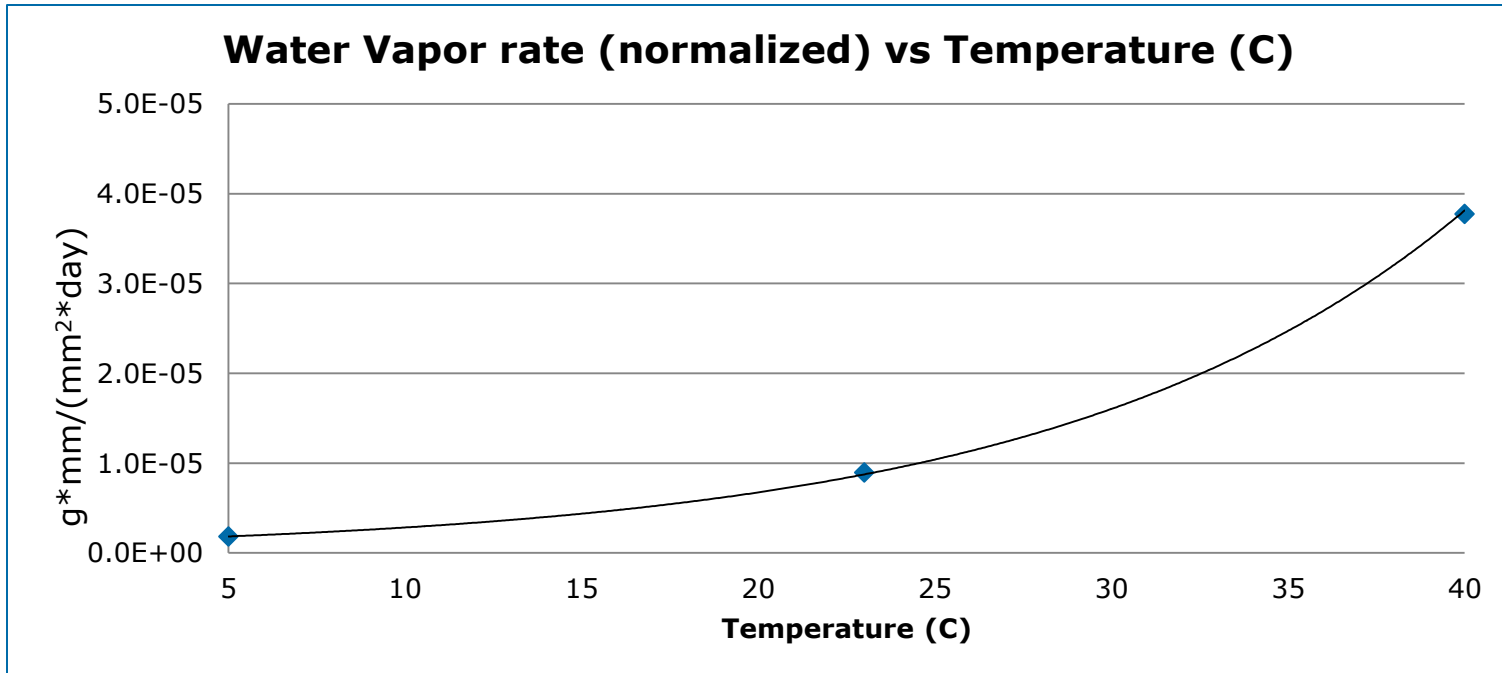
Conclusion: The molded plastic process was optimized by Catalent to reduce the temperature of the solution (0.5 mL fill) close to 40° C at time of fill and the assumption is that the temperature of the solution would fall steadily after the units come out of the fill suite and packaged under ambient conditions.

Oxygen and Carbon Dioxide Gas Permeation Data



Conclusion: Carbon Dioxide has higher permeation rate relative to Oxygen because Carbon Dioxide partitions into the container to a greater extent relative to Oxygen.

Water- Vapor Transmission Rates



Conclusion: Higher the Temperature, greater the water vapor permeation rates



Case – Study: Compatibility assessment of a model monoclonal antibody formulation with Advasept™ and Glass container system



DEVELOPMENT



DELIVERY



SUPPLY

more products. better treatments. reliably supplied.™

Monoclonal Antibody Formulation Recipe + Preparation

Table 1: Model mAb Formulation

Components	Amount (mg/mL)
Model mAb (Mol. Wt. 144 kDa)	10
Polysorbate 80	0.7
Sodium Citrate	6.5
Sodium Chloride	9.0
pH	6.5

The monoclonal antibody formulation was filtered with a 0.2 micron Nalgene filter unit and filled in glass vials into which uncoated stoppers were placed. The mAb formulation was filtered into a sterile bag and shipped to Blow-Fill-Seal (BFS) manufacturing facility. The formulation was aseptically transferred in BFS Advasept™ stoppered vials.

mAb Compatibility Assessments (Glass/Advasept Vial formats)

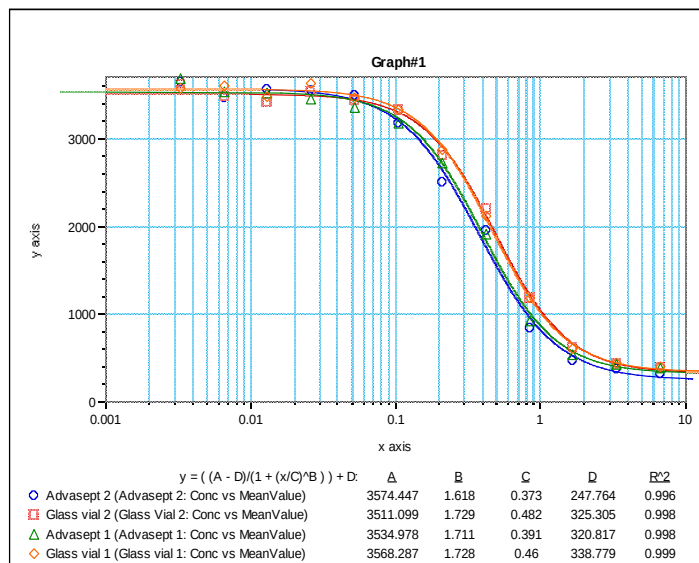
Parameter	Method	Target Range
pH	USP 791	6.5 $\pm$ 0.3
Appearance	Visual Inspection	Report Results
Potency	UV (280 nm)	T=0 $\pm$ 10%
	Activity	Report EC ₅₀
Stability	Size-Exclusion Chromatography (SEC)	Report % Monomer and % High and low molecular weights
	Nanoparticle Tracking Analysis (NTA) ⁵	Report sub-micron particle size analysis
	Sodium Dodecyl Sulfate-Polyacrylamide Gel Electrophoresis (SDS-PAGE)	Report % Area (HC +LC)
	Capillary Isoelectric focusing (cIEF)	pI (% of each peak)
	Peptide Mapping ²	% Chemical Modification
Leachables ³	Polar Leachables (HPLC-UV)	Report Irganox 1010 levels
	Semi-Volatile Leachables (GC-MS)	Report Aromatic Hydrocarbon levels
	Volatile Leachables GC-FID	Report Volatile Leachable levels ⁴
	Metals (ICP-MS)	Report all metals except Na, I
Bacterial Endotoxin ¹	USP 85	Report Results
Bioburden ¹	USP 61	<10 CFU/mL
¹ only performed at 0 time-interval		
² Only performed at 4 month time-interval		
³ Only performed at 9M time-interval		
⁴ Isopropyl alcohol, Methyl Ethyl Ketone, Trichloroethylene, Hexane, 2-methyl pentane, 3-methyl pentane, Methylcyclopentane, Cyclohexane and Heptane		
⁵ Performed on 12 M samples		

Results 1 -Potency

Potency:

- a) **UV:** The UV data indicates no apparent change in potency for both vial formats upon pre and post-fill and upon storage to 9 months at 5°C were within target range.
- b) **Complement Dependent Cytotoxic Assay:** The potency was determined using responsive cell line in a complement dependent cytotoxic assay using a fluorescence read out. Comparison of a dilution series with standard, formulation in glass and the Advasept™ vial were generated (Figure 1).

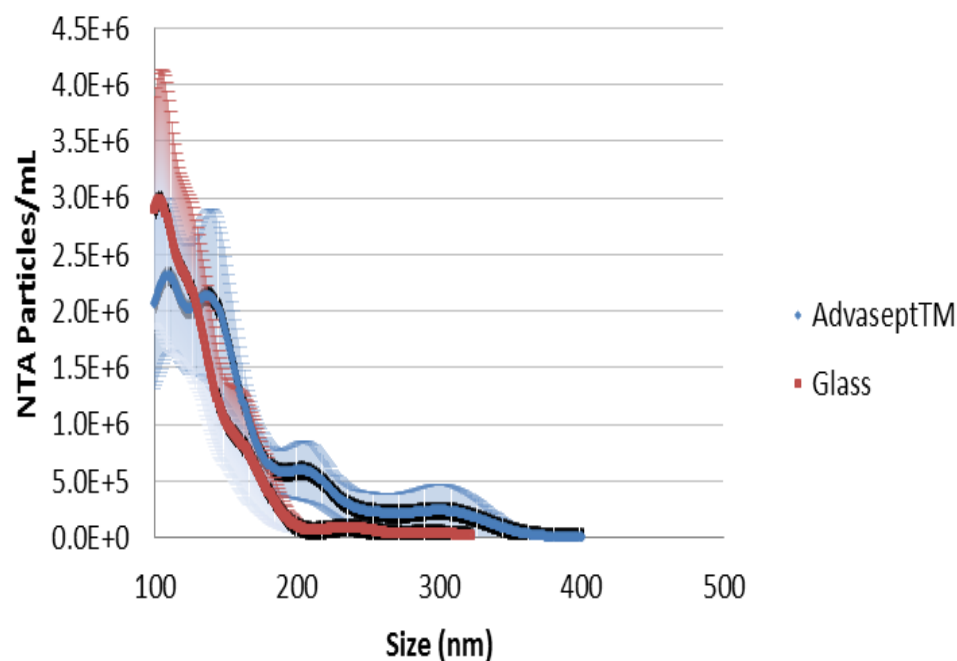
Figure 1: Dose Response Curve for model mAb formulation



Conclusions: The activity data shows comparable potency values between the Glass and Advasept™ vials upon storage to 24M/5° C (All data not shown).

Result 3 -Nanoparticle Tracking Analysis (NTA)

NTA Rightway Particles in Glass/Advasept™

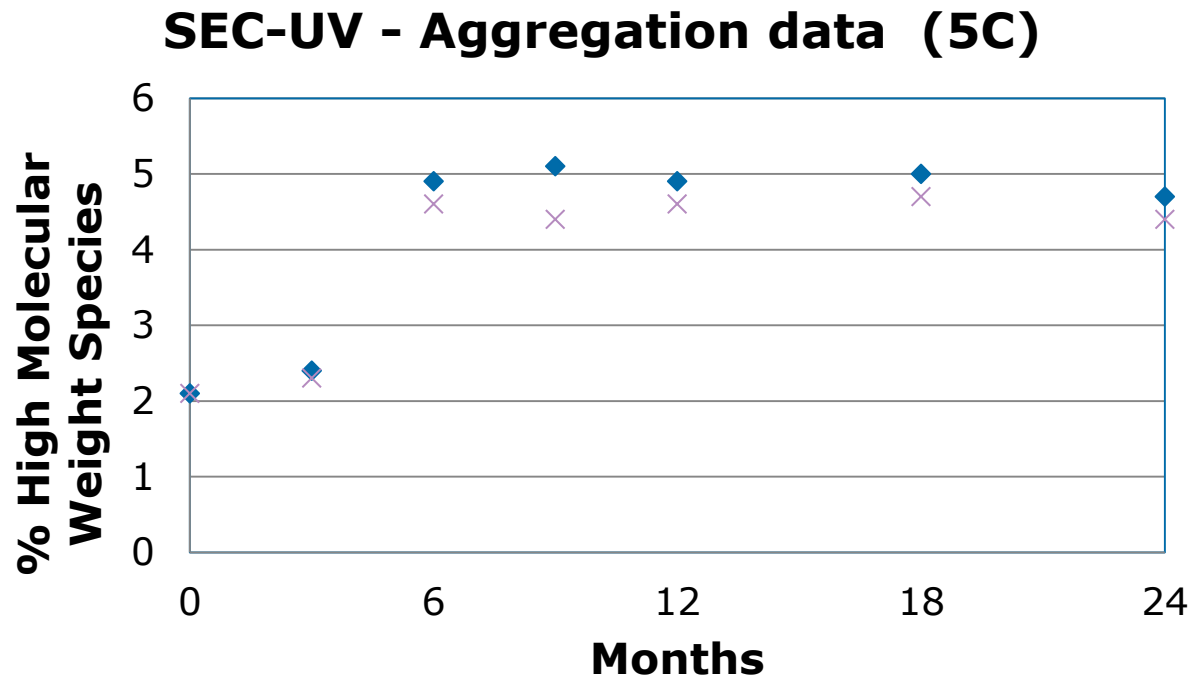


Particles	Advasept™	Glass Vial
D ₅₀	126 ± 17.5	94 ± 10.1 nm
D ₉₀	205 ± 35 nm	144 ± 13.4 nm
Total Concentration (particles/frame)	13.95 ± 3.2	13.47 ± 3.0
Total Concentration (particles/mL)	2.83e ⁸ ± 6.5e ⁷	2.74e ⁸ ± 6.1e ⁷

Mean sub-micron particle count data
(Nanoparticle Tracking Analysis)
for Monoclonal antibody formulations stored in
Glass and Advasept™ vials for 12M/5°C.

Conclusions: Slightly higher size mode of aggregates were observed in Advasept™ than in the glass vials, however, the total number of particles are statistically comparable between the two container closure systems.

Result 2 SEC-UV Aggregation Data



Conclusion: *Stability data indicates higher levels of high molecular weight species in Glass relative to Advasept™ vial.*

Results 4-Peptide Mapping Results

Procedure: Aged Glass and Advasept™ vials (4M/5°C) were subjected to peptide mapping by initially denaturing and reduction of the mAb with DTT, alkylation with Iodoacetamide, followed by clean-up on a column, and digestion with Trypsin and ASP-N. The peptides were separated on a UPLC column and UV and MS detectors were employed. (Chromatograms provided in the appendix section).

Type	Sample	% Modification
Oxidation	Glass Vial	10.8
	Advasept™ Vial	4.5
Deamidation	Glass Vial	7.2
	Advasept™ Vial	7.6

Conclusions: The oxidation levels were higher in Glass relative to Advasept™ vials and could be attributed to mAb's surface interaction with glass surface causing more oxidation than in plastic Advasept™ vials.

Results 5 (pH, Appearance, cIEF and SDS-PAGE)

All other stability test results for parameters listed below were comparable between Glass and Advasept™ vial formats.

- 1. pH*
- 2. Appearance*
- 3. cIEF*
- 4. SDS-PAGE*

Bioburden results performed at t=0 for both vial configurations were below <10 CFU/mL.

Results 6 (Leachable Data Analysis)

The monoclonal antibody formulation samples stored for 9M at 5° C were analyzed for various leachables listed below:

1.Volatile: GC-FID

2.Semi-Volatile: GC-MS

3.Polar: HPLC-UV

4.Metal leachables: ICP-MS

Results: *The leachable data indicates comparable leachable profile for monoclonal antibody formulations stored in Glass and Advasept™ vials except for Isopropyl alcohol. The Isopropyl alcohol content was determined to be higher in Glass vial (14 mg/mL) relative to Advasept™ vial (1.2 mg/mL) and was attributed to cleaning procedure used for glass vials prior filling of the monoclonal antibody formulation.*

Conclusions

1. This study confirms compatibility of monoclonal antibody formulation in glass and Advasept™ vial demonstrating plastic BFS vial suitability for protein therapeutics.
2. Stability data indicates higher levels of high molecular weight species in Glass relative to Advasept™ vial.
3. Affinity data indicates comparable potency levels in Glass and Advasept™ vial.
4. Leachable data indicates comparable leachable profiles in Glass and Advasept™ vial.
5. No significant differences were identified between Glass and Advasept™ vials over the 9 months analyzed to date.

ACKNOWLEDGEMENTS

Catalent- Woodstock

- 1. Waiken Wong, Ph.D.**
- 2. Bill Hartzel**
- 3. Natasha Hults**
- 4. Kay Schmidt**

Catalent – Madison

- 1. Gregory Bleck, Ph.D.**
- 2. Ian J. Collins, Ph.D.**
- 3. Process Development Team**

Catalent – Kansas City

- 1. Vincy Abraham, Ph.D.**

Catalent - RTP

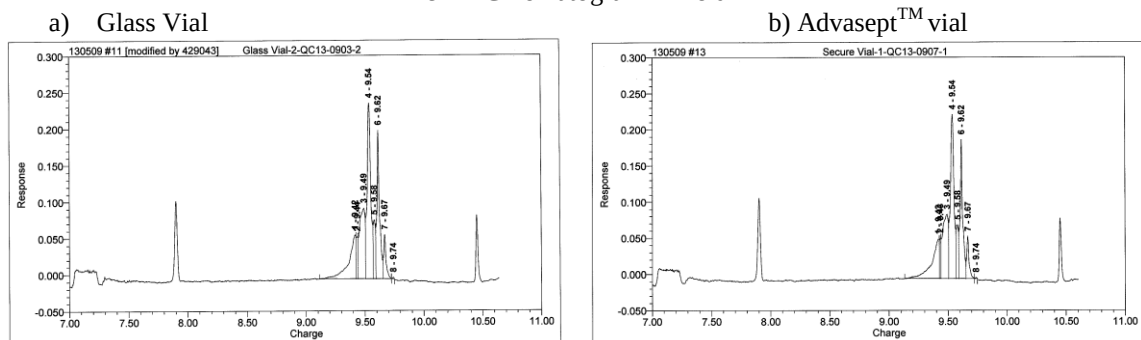
- 1. Thomas Luntz**
- 2. Courtney Jones**
- 3. James Mclean**
- 4. Vicki Wards**

QUESTIONS ???

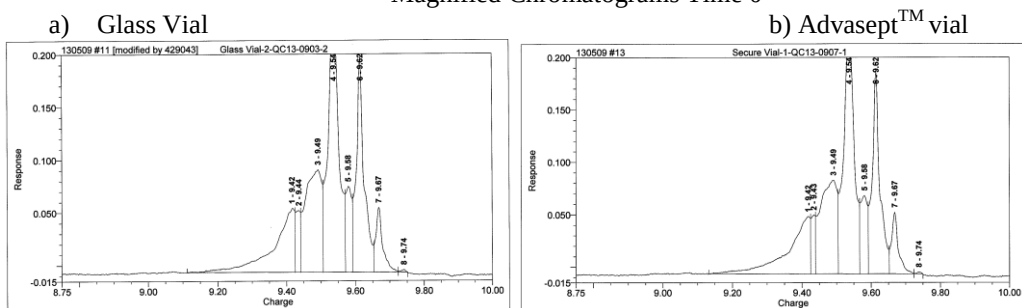
APPENDIX

cIEF Analysis

cIEF Chromatogram Time 0



Magnified Chromatograms Time 0



The chromatograms showed no comparable charge distribution between the Glass and Advasept™ vial.

Impact of Elevated Temperature of the Molded Plastic during filling on the stability of biologic formulation

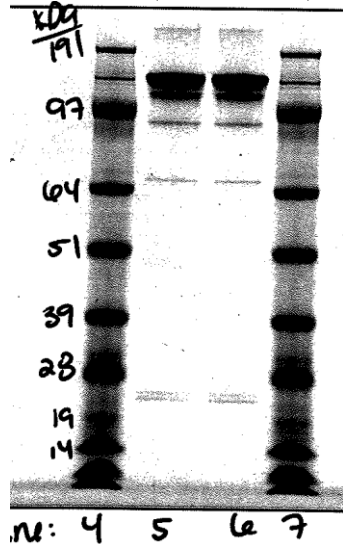
		T=0 data								
Parameters	Bulk Solution			Glass Vial			Advasept™ Vial			
pH	6.4			6.6			6.6			
Potency (UV) (mg/mL)	10.8			10.8			10.9			
SEC	2.2% High Mol. Wt. 97.8% Monomer			2.1% High Mol. Wt 97.9% Monomer			2.1% High Mol. Wt. 97.9% Monomer			
SDS	Similar Profile (Reduced/Non Reduced) Figures – Appendix section									
cIEF	Major PIs (%)* pI Range 9.38-9.65 (7 peaks)			Major PIs (%) pI Range 9.42-9.74 (8 peaks)			Major PIs (%) pI Range 9.42-9.74 (8 peaks)			
cIEF (peaks)	Peak	pI	% Area	Peak	pI	% Area	Peak	pI	% Area	
	1	9.38	11.0	1	9.42	15.9	1	9.42	15.8	
	2	9.40	14.0	2	9.44	3.2	2	9.44	3.1	
	3	9.45	11.9	3	9.49	18.8	3	9.49	18.6	
	4	9.50	34.0	4	9.54	31.8	4	9.54	31.9	
	5	9.56	3.49	5	9.58	6.2	5	9.58	6.2	
	6	9.59	20.7	6	9.62	18.9	6	9.62	19.1	
	7	9.65	5.0	7	9.67	5.1	7	9.67	5.3	
			8	9.74	0.1	8	9.74	0.2		

Conclusion: Data from stability indicating parameters between the bulk mAb, glass and Advsept™ vial drug product indicates that the BFS process is amenable to Biologics.

**analysis occurred at separate time using different cIEF cartridge which may explain slight differences and possible poor resolution of peak 8 which is only 0.1% of the total area. Peak profiles look similar.*

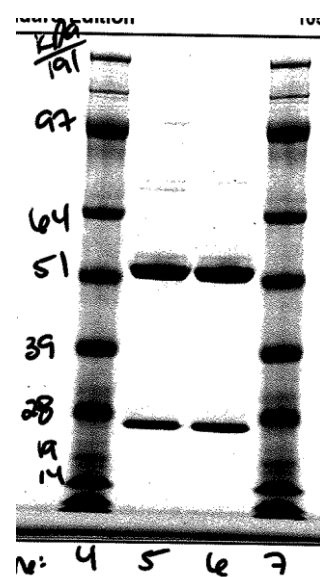
SDS – PAGE Analysis

SDS-PAGE (Non-Reduced)



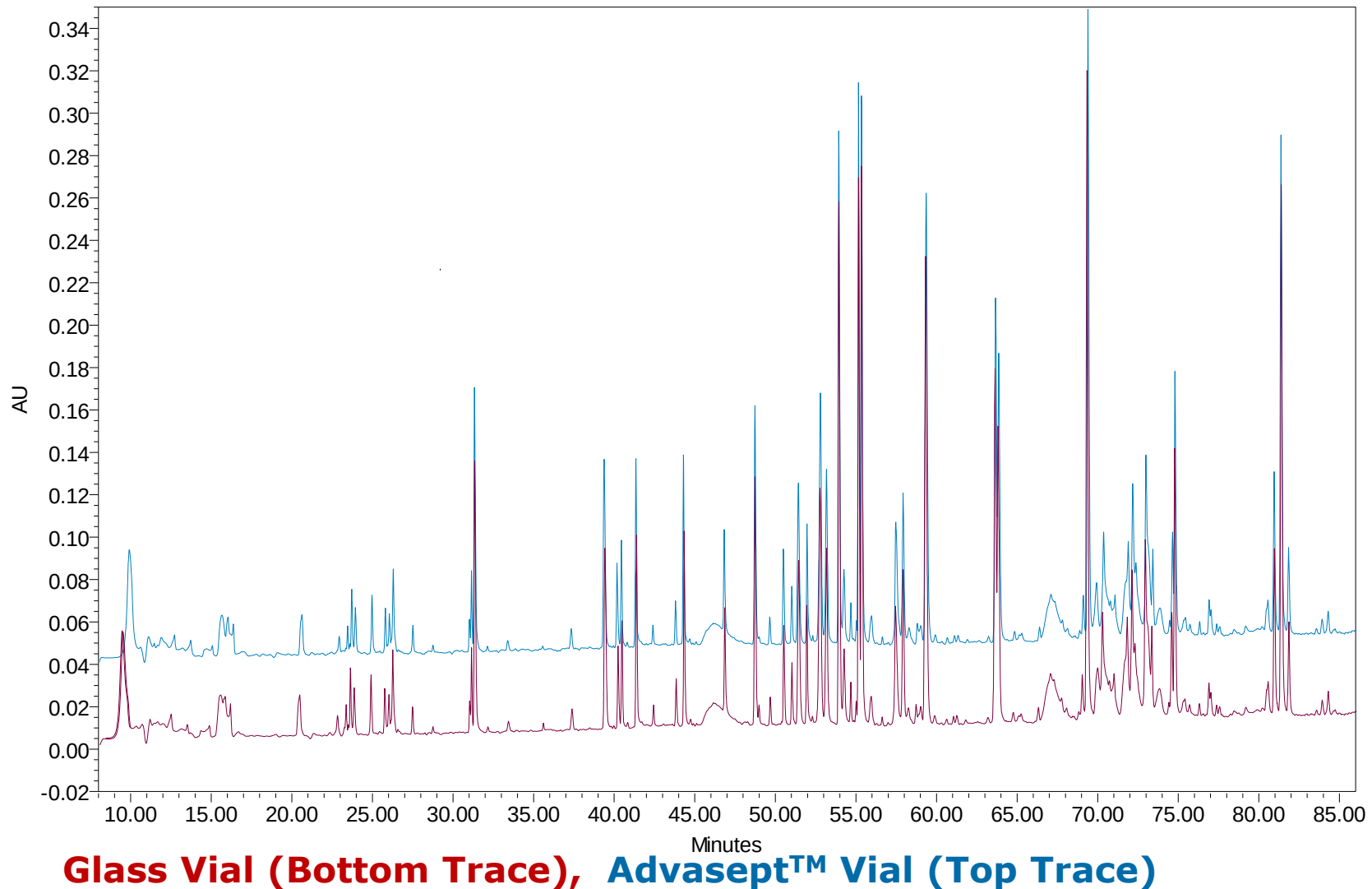
Lane 5 – Glass Vial
Lane 6 – Advasept™ Vial

SDS-PAGE (Reduced)

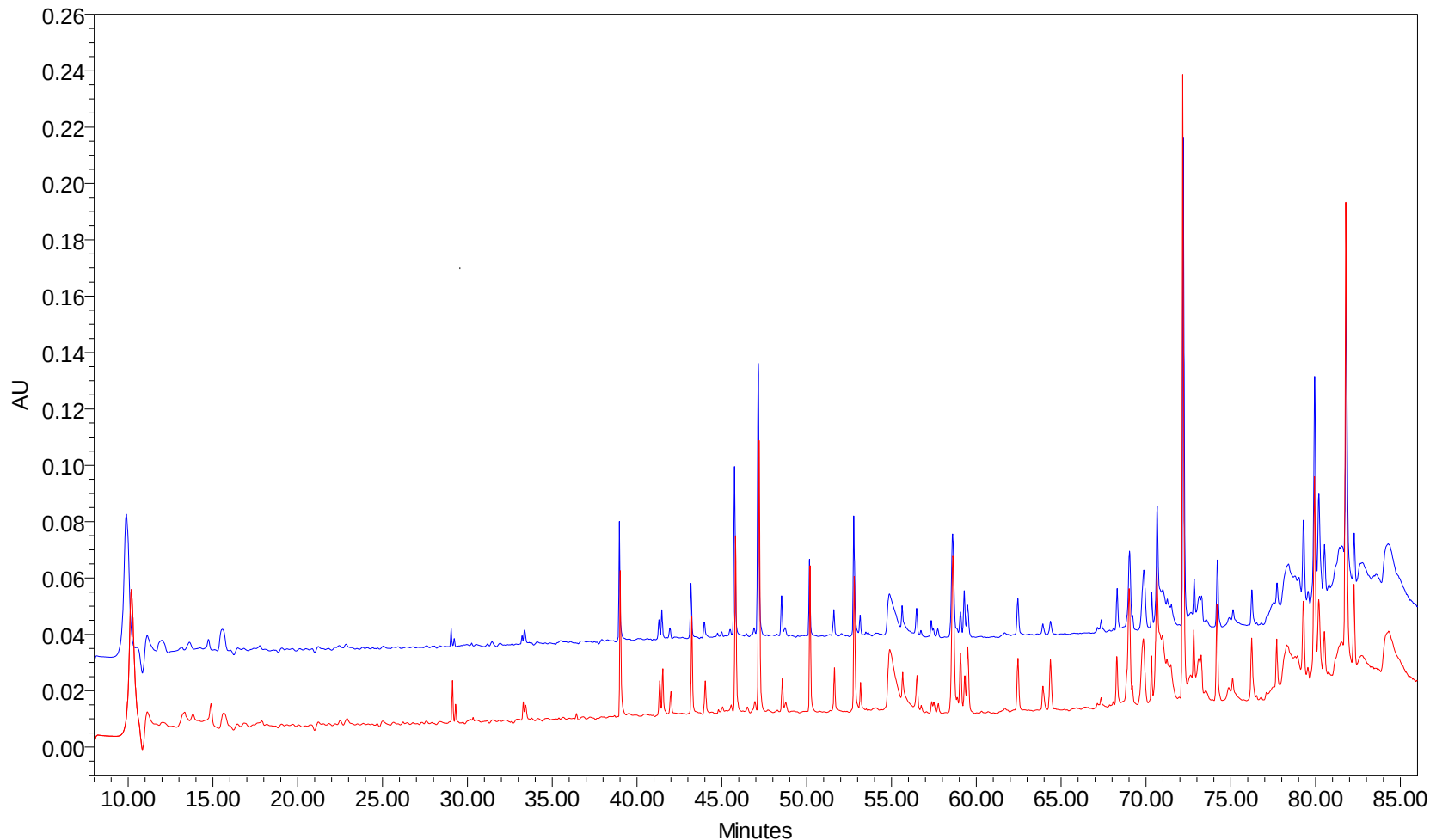


Lane 5 – Glass Vial
Lane 6 – Advasept™ Vial

Magnified UPLC/UV Chromatograms of Trypsin Digested Monoclonal Antibody



Magnified UPLC/UV Chromatograms of ASP-N Digested Monoclonal Antibody



Glass Vial (Bottom Trace), Advasept™ Vial (Top Trace)



Let us meet again..

**We welcome you all to our future conferences
of OMICS International**

**2nd International Conference and Expo
on**

Parenterals and Injectables

On

October 24-26, 2016 at Istanbul, Turkey

**[http://parenterals-
injectables.pharmaceuticalconferences.com/](http://parenterals-injectables.pharmaceuticalconferences.com/)**