Protein Conformational Arrays for theraputic mAb Development.

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xMap Connect, Amsterdam, 2018

Topics Covered Today

- The need for new technologies?
- Technology Development.
- Bridging Studies of PCA : from ELISA to xMAP.
- Study mAb HOS and Immunogenicity Correlation with PCA.
- Conclusions.

1.The need for a novel Technology for Protein Conformational Analysis?

Novel Therapeutic Mab Development: The Market*.

First approved therapeutic monoclonal antibody product in 1986 (Orthoclone, Kidney Disease)
By November 10, 2014, 47 monoclonal antibody with EU/USA Clearance.
At current approval rates by 2020 70 expected with a cap of $125bn

* Ecker et al, The Theraputic Monoclonal Antibody Market. mAbs 7:1, 9--14; January/February 2015;

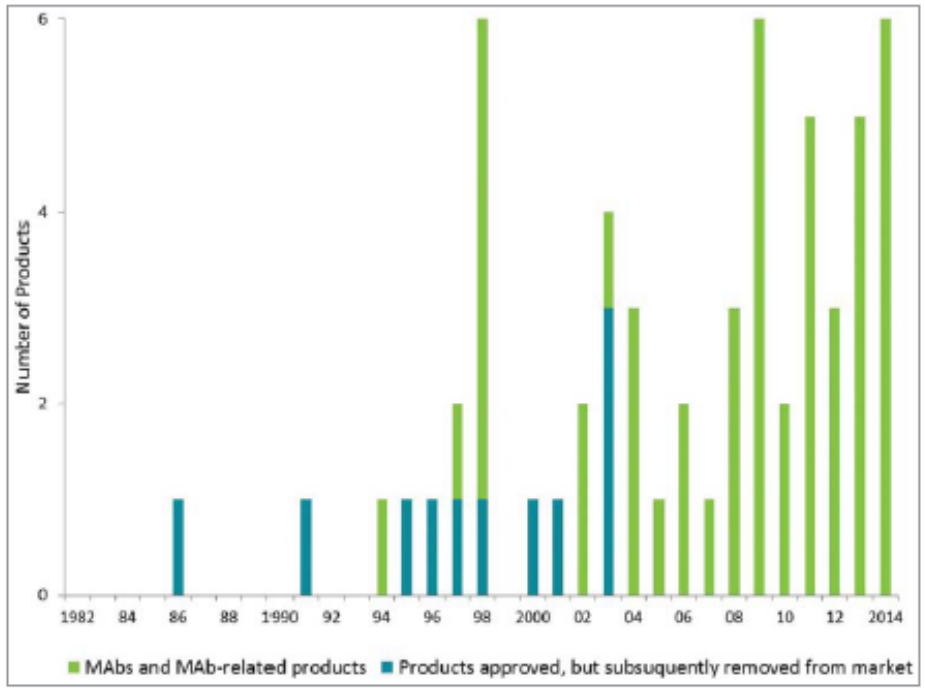


Figure 1. Annual approvals of monoclonal antibody products.[3,4] The number of monoclonal antibody products first approved for commercial sale in the US or Europe each year since 1982 is shown. The totals include all monoclonal antibody and antibody-related products. Products approved but subsequently removed from the market are denoted in blue; products currently marketed are denoted in green. 2014 total is as of November 10, 2014.

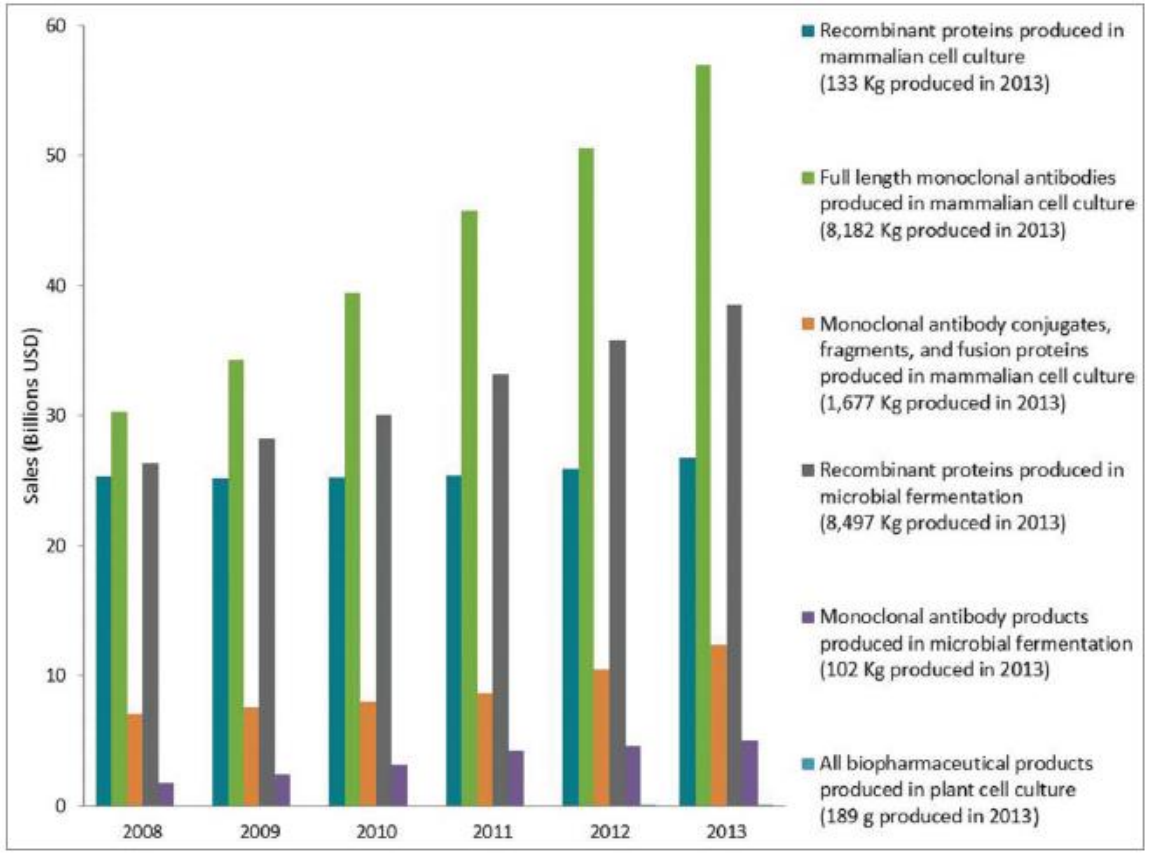
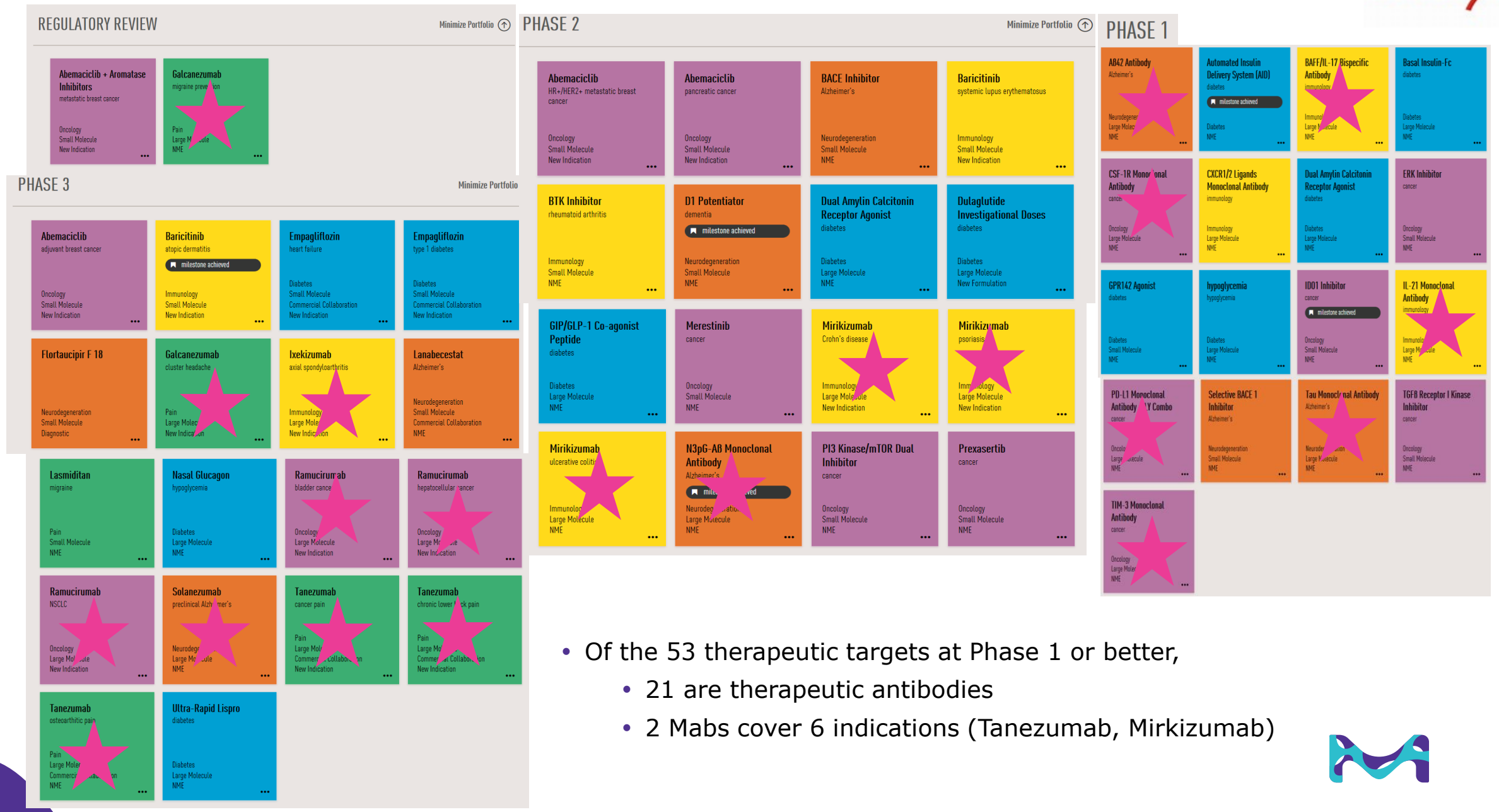


Figure 2. Sales of biopharmaceutical products by product type. Total annual sales of biopharmaceutical products are shown as a function of product type. Note that recombinant proteins produced by microbial fermentation include recombinant human insulin products which represent nearly 50% of the sales and >90% of the material produced in this category.

Big Pharma Example: Eli Lilly Pipeline https://www.lilly.com/discovery/pipeline



- Of the 53 therapeutic targets at Phase 1 or better,
  - 21 are therapeutic antibodies
  - 2 Mabs cover 6 indications (Tanezumab, Mirkizumab)

Approved Therapeutic Mabs to 2015

Table 1. Marketed therapeutic monoclonal antibody products

Brand name (INN)	Original BLA/MAA Applicant	Company Reporting EU Sales	Year of First Approval	2013 Global Sales ($M)[a]
Abthrax (raxibacumab)	Human Genome Sciences	N/A[b]	2012	23
Actemra (tocilizumab)	Roche	Roche	2009	1,119
Adcetris[c] (brentuximab vedotin)	Seattle Genetics	Takeda Pharmaceutical Co.	2011	253
Alprolix[d] (Factor IX Fc fusion protein)	Biogen Idec	N/A	2014	NoM[e]
Arcalyst[f] (rilonacept)	Regeneron Pharmaceuticals	N/A	2008	17
Arzerra (ofatumumab)	GlaxoSmithKline	GlaxoSmithKline	2009	117
Avastin (bevacizumab)	Genentech	Roche	2004	6,748
Benlysta (belimumab)	Human Genome Sciences	GlaxoSmithKline	2011	228
Cimzia[g] (certolizumab pegol)	UCB	UCB	2008	789
Cyramza (ramucirumab)	Eli Lilly and Co.	N/A	2014	NoM[e]
Eloctate[h] (Factor VIII Fc fusion protein)	Biogen Idec	N/A	2014	NoM[e]
Enbrel[i] (etanercept)	Immunex	Pfizer	1998	8,325
Entyvio (vedolizumab)	Takeda Pharmaceuticals U.S.A., Inc	Takeda Pharmaceutical Co.	2014	NoM[e]
Erbitux (cetuximab)	ImClone Systems	Merck KGaA	2004	1,926
Eylea[j] (aflibercept)	Regeneron Pharmaceuticals	Bayer Healthcare Pharmaceuticals	2011	1,851
Gazyva (obinutuzumab)	Genentech	Roche	2013	3
Herceptin (trastuzumab)	Genentech	Roche	1998	6,559
Humira (adalimumab)	Abbott Laboratories	AbbVie	2002	10,659
Ilaris (canakinumab)	Novartis Pharmaceuticals	Novartis Pharmaceuticals	2009	119
Inflectra[k l] (infliximab [biosimilar])	Hospira	Hospira	2013	<1[m]
Kadcyla[n] (ado-trastuzumab emtansine)	Genentech	Roche	2013	252
Keytruda (pembrolizumab)	Merck & Co.	N/A	2014	NoM[e]
Lemtrada (alemtuzumab)	Genzyme Therapeutics	Sanofi	2013	3
Lucentis[o] (ranibizumab)	Genentech	Novartis Pharmaceuticals	2006	4,205
Nplate[p] (romiplostim)	Amgen	Amgen	2008	427
Nulojix[q] (belatacept)	Bristol-Myers Squibb	Bristol-Myers Squibb	2011	26
Orencia[r] (abatacept)	Bristol-Myers Squibb	Bristol-Myers Squibb	2005	1,444
Perjeta (pertuzumab)	Genentech	Roche	2012	352
Prolia[s] (denosumab)	Amgen	GlaxoSmithKline	2011	824
Remicade (infliximab)	Centocor	Merck & Co.	1998	8,944
Removab[t] (catumaxomab)	Fresenius Biotech	NeoPharm Group	2009	5
Remsima[k l] (infliximab [biosimilar])	Celltrion	Celltrion	2013	<1[m]
ReoPro[u] (abciximab)	Centocor	N/A	1994	127
Rituxan (rituximab)	Genentech	Roche	1997	7,500
Simponi/ Simponi Aria (golimumab)	Centocor Ortho Biotech	Merck & Co.	2009	1,432
Simulect (basiliximab)	Novartis Pharmaceuticals	Novartis Pharmaceuticals	1998	30[v]
Soliris (eculizumab)	Alexion Pharmaceuticals	Alexion Pharmaceuticals	2007	1,551
Stelara (ustekinumab)	Janssen-Cilag International	Johnson & Johnson	2009	1,504
Sylvant (siltuximab)	Janssen Biotech	Johnson & Johnson	2014	NoM[e]
Synagis (palivizumab)	Abbott Laboratories	Abbvie	1998	1,887
Tysabri (natalizumab)	Biogen Idec	Biogen Idec	2004	1,527
Vectibix (panitumumab)	Amgen	Amgen	2006	389
Xgeva[s] (denosumab)	Amgen	Amgen	2010	1,030

- Most of these are novel Mabs.
- However biosimilars are starting to be approved.
- FDA approved 1st full length mAb biosimilar in 2016.
- EMA approval was in 2013.

Therapeutic Mab Work Flow

Protein Journey

Originator Mab
Discovery-Non Stable Cell Line

Biosimilar
Reference Standard

MFG Cell Line*
- Cell Line Development
- Media Optimization

R=Protein Concentration

"Totality of Evidence"

Structural Analytics
- CD Spectrum
- Size Exclusion Chromatography (SEC)
- Analytical Ultracentrifugation (AUC)
- Non-denaturing Electrophoresis
- Hydrogen/deuterium exchange (HDX)
- NMR

R=Similarity to Reference

Sample Analysis
- Nonclinical
- PK
- PD
- Clinical Safety
- Immunogenicity

R= Safety/Efficacy

Manufacturing Upscale (Downstream)

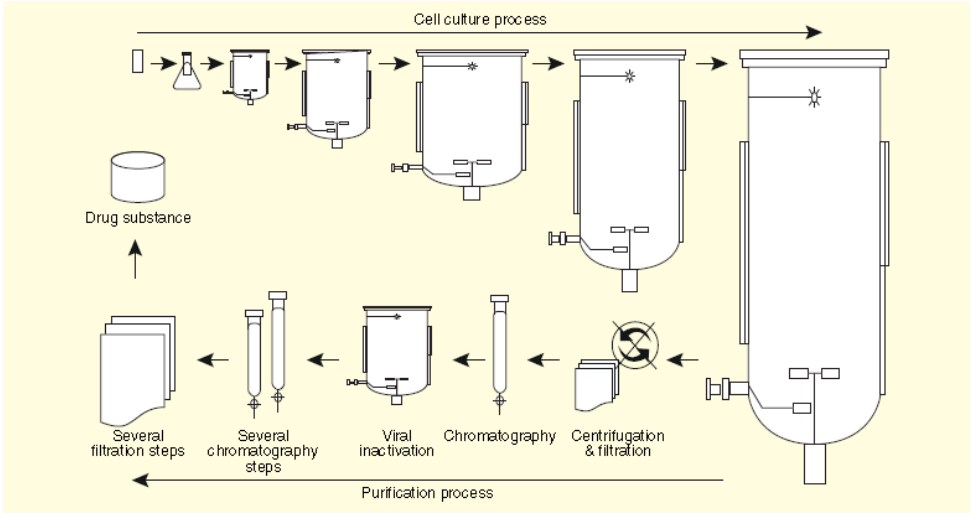


Figure 1. Overview of the manufacturing process of a biologic in a mammalian cell culture system. The manufacturing process consists of a cell culture process and a purification process. The cell culture process is initiated by expansion of a single vial of cell stock to culture flask, after which it is sequentially subcultured to larger bioreactors. In this process, optimized cell culture conditions such as temperature, agitation rate, osmolality, pH, concentration of CO_2 and glucose concentration are tightly maintained, as these conditions are critical to the quality of biologics. The supernatants are harvested and further purified through several steps of chromatography, filtration and viral inactivation in the purification process, which also have potential to influence the quality of biologics.

R=Similarity to Reference

Formulation
Upscale
Bulk Processing

Finished Good
Packaging
Distribution

*** Cell Line.**
- Work with a well established cell line (avoid Allotypic variation)
- Optimize media to avoid changes in Glycosylation.

This may be applied to ANY therapeutic protein

The Problem:

Quality Considerations in Demonstrating Biosimilarity of a Therapeutic Protein Product to a Reference Product. Guidance for Industry. FDA, April 2015.

"The three dimensional conformation of a protein is an important factor in its biological function. Protein generally exhibit complex three-dimensional conformations (tertiary structure and, in some cases, quaternary structure) due to their large size and the rotational characteristics of protein alpha carbons. The resulting flexibility enables dynamic, but subtle, changes in protein conformation over time, some of which may be absolutely required for functional activity." "…… at the same time, a protein's three-dimensional conformation can often be difficult to define precisely using current physiochemical analytical technology."

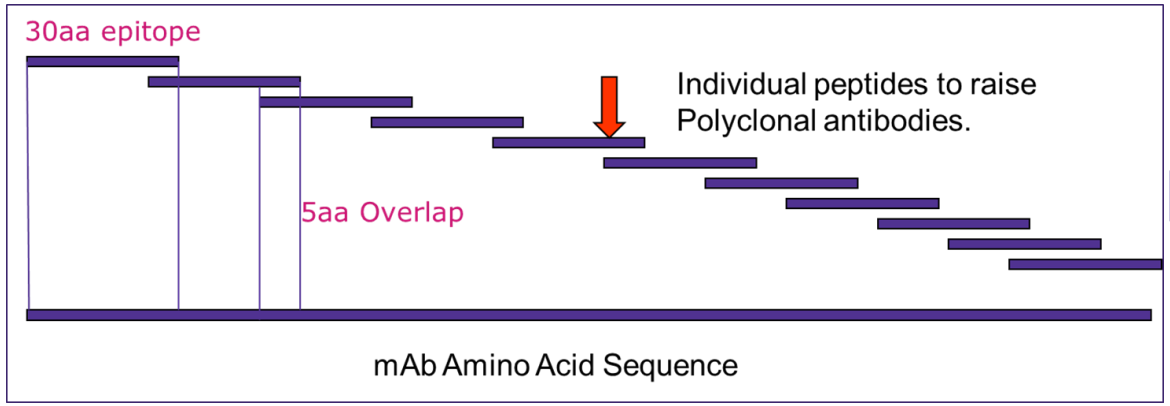
Current Technologies for Conformational Analysis

HOS Technologies	Principle	Pros	Cons
CD	Peptide bond and aromatic amino acid environment	Easy to use, low cost	Low sensitivity, average of the whole measured population.
FTIR	Peptide bonds	Easy to use, low cost	Low sensitivity, average of the whole measured population.
PCA	Epitope recognition by antibodies.	Easy to use, cGMP friendly, systematic, high sensitivity and throughput.	New to the market. Intermediate cost level.
HDX-MS	Hydrogen-Deuterium exchange in the amide group of protein surface	High resolution, well-established applications.	High cost, needs special instrument and training, low throughput.
HRF-MS	Free radical labeling of protein surface hydroxyl groups	High resolution.	High cost, needs special instrument and training, low throughput.
Bioassay	Target recognition	Well-established. cGMP friendly.	Low resolution
X-Ray	Atom diffraction	High resolution.	High cost, Low throughput, not suitable for routine testing.
DLS	Aggregate and multimer light scattering	Low cost, well-established.	Low sensitivity, average of the whole measured population.
NMR	Nuclei spin and charge	High resolution, well-established.	Low throughput, needs special instrument and training. High cost. For small proteins.
Fluorescence	Aromatic amino acid environment	Low cost, well-established.	Low sensitivity, average of the whole measured population.

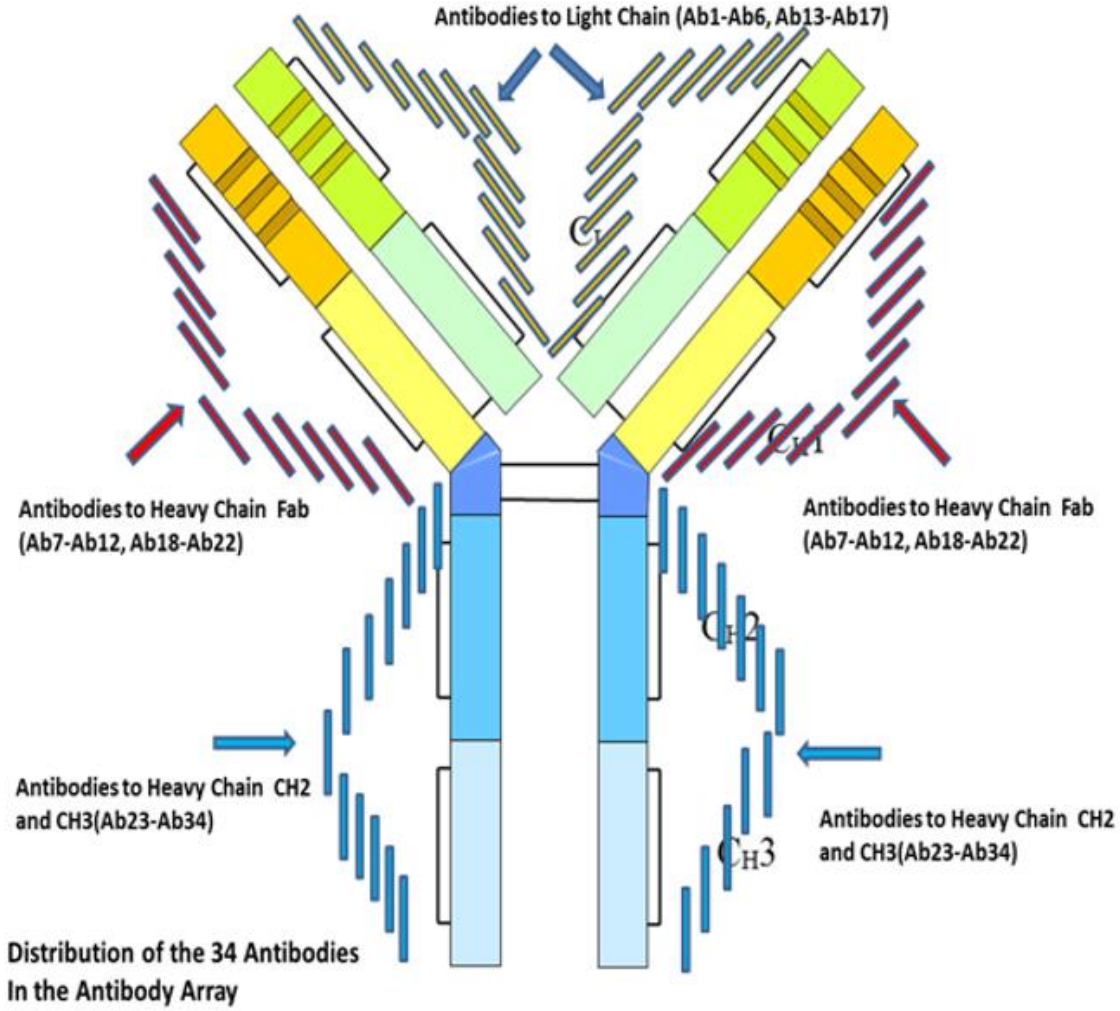
2. Technology Development

PCA Technology Covers the Whole mAb

- **Polyclonal Antibodies (Pab) are raised against 30 amino acid peptides from the amino acid sequence of target therapeutic Mab.**



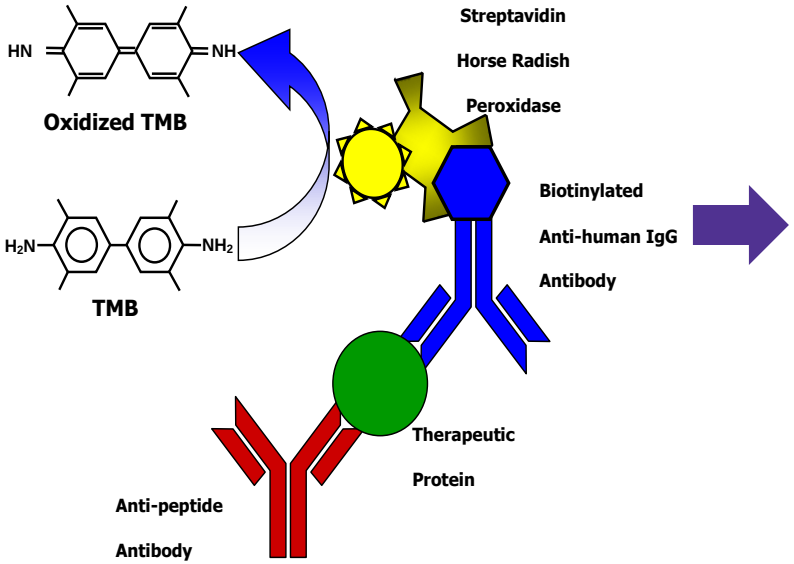
- **Selected Pab's are used to create an array against the structure of the Mab. With a reference molecule providing a figerprint of the properly folded Mab.**

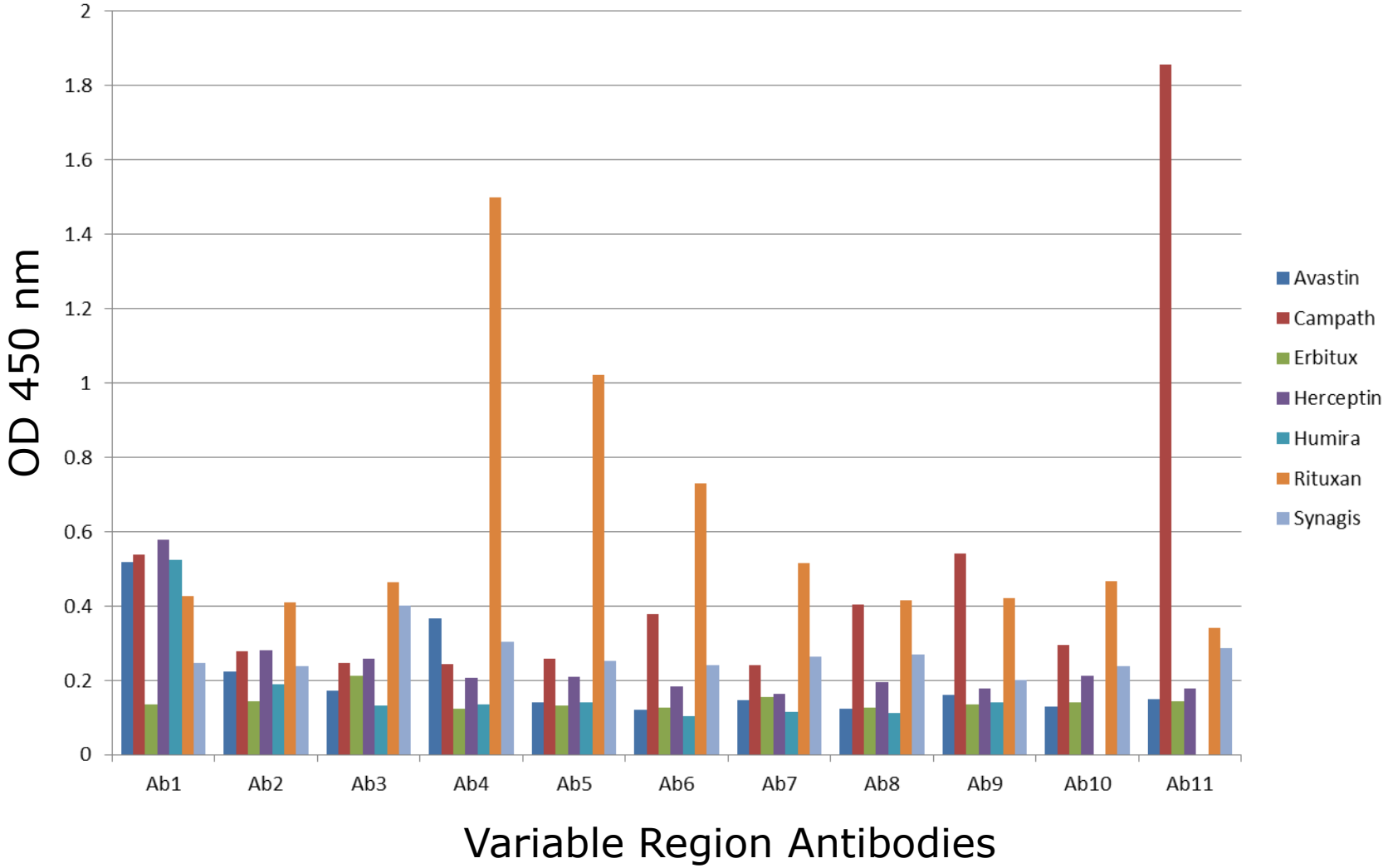


Distribution of the 34 pAb: pAb 1-12 (variable region); pAb 13-34 (constant region)

PCA ELISA's

The initial product offering consist of Sandwich ELISA based arrays, allowing the generation of unique therapeutic Mab signatures.



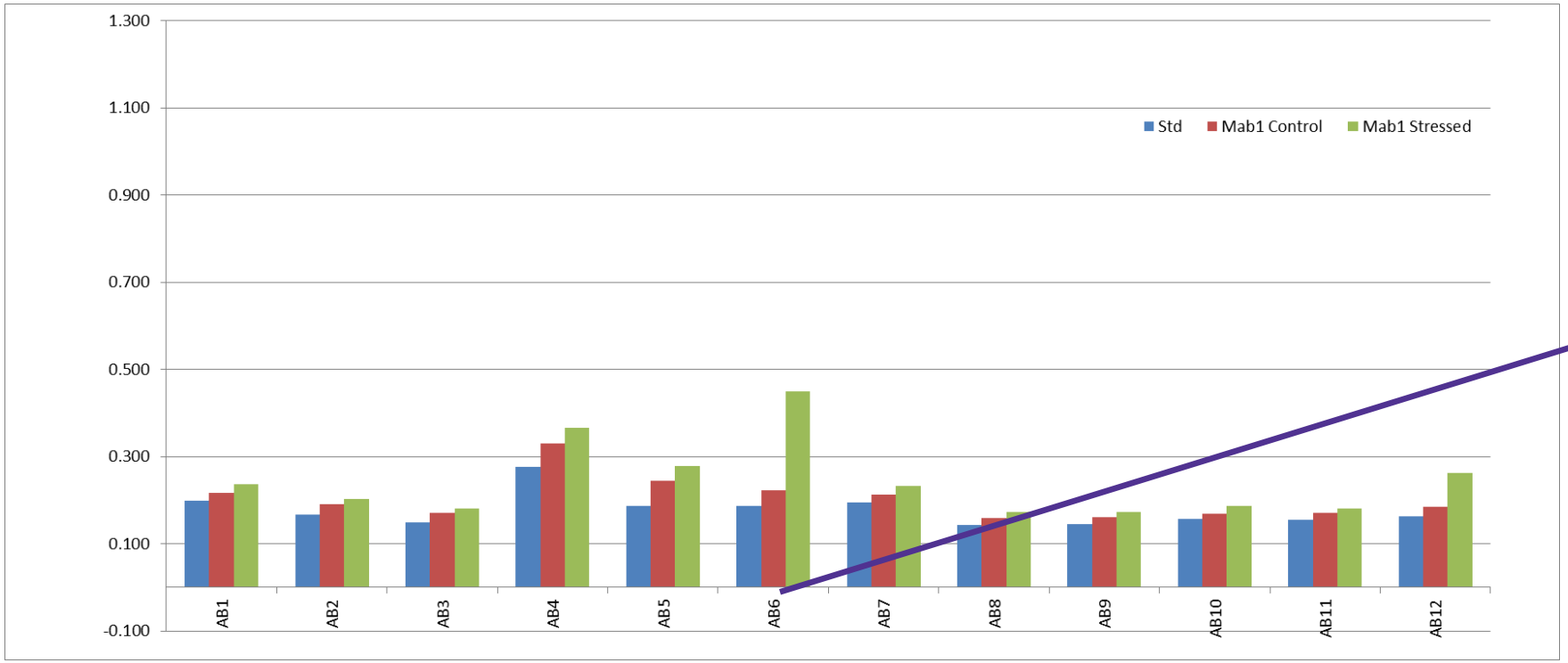


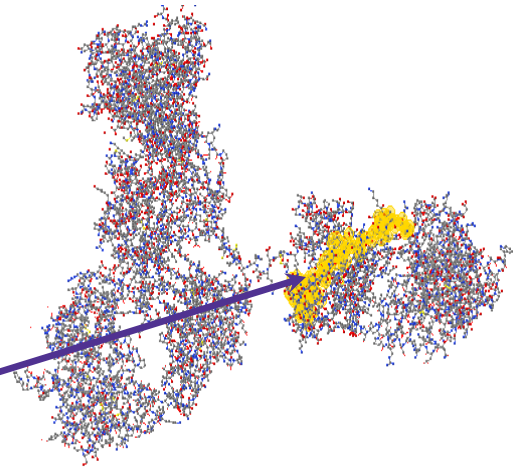
Protein Conformational Array (PCA): A Multifaceted "Fingerprint-like" Analytical Technology for Biosimilarity Evaluation

Attributes Monitored	PCA ELISA Detection	Sensitivity	Molecular Resolution
Temperature Stress	Yes	0.1% (5 ng impurity in 5 µg testing sample)	Epitope-based, 3-6 Amino Acids
Low pH	Yes	High	Epitopes
High pH	Yes	High	Epitopes
Oxidation	Yes	High	Epitopes
Glycosylation	Yes	High	Epitopes
Aggregation	Yes	High	Epitopes
Bioassay Difference	Yes	High	Epitopes
Light Stress	Yes	High	Epitopes

Case study: Correlation Between Conformation and Bioassay in Stability Testing (Novel mAb)

The most significant difference in the variable region was seen at Ab6 suggesting a correlation between this site and the decrease in bioactivity (the more unfolding the higher the signal)

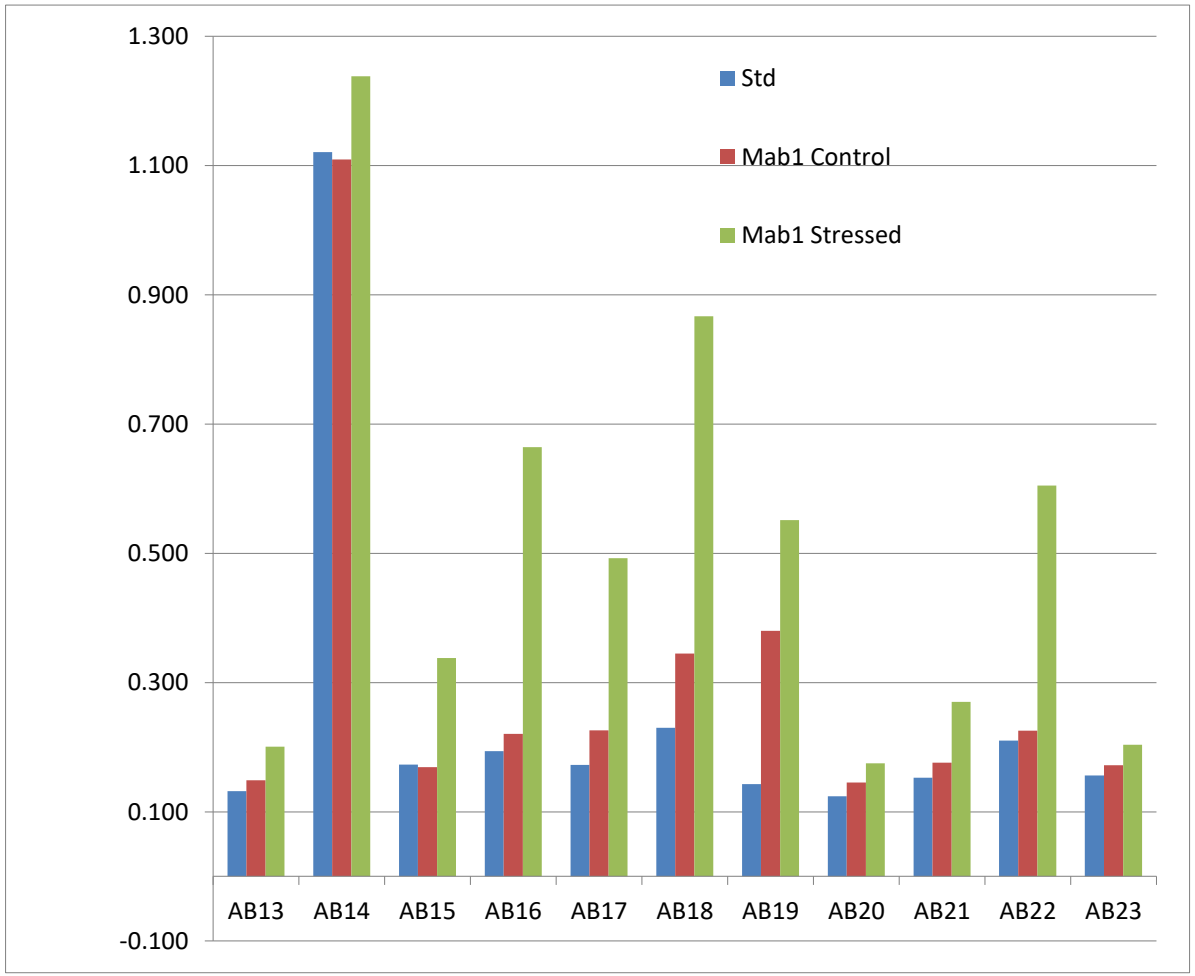


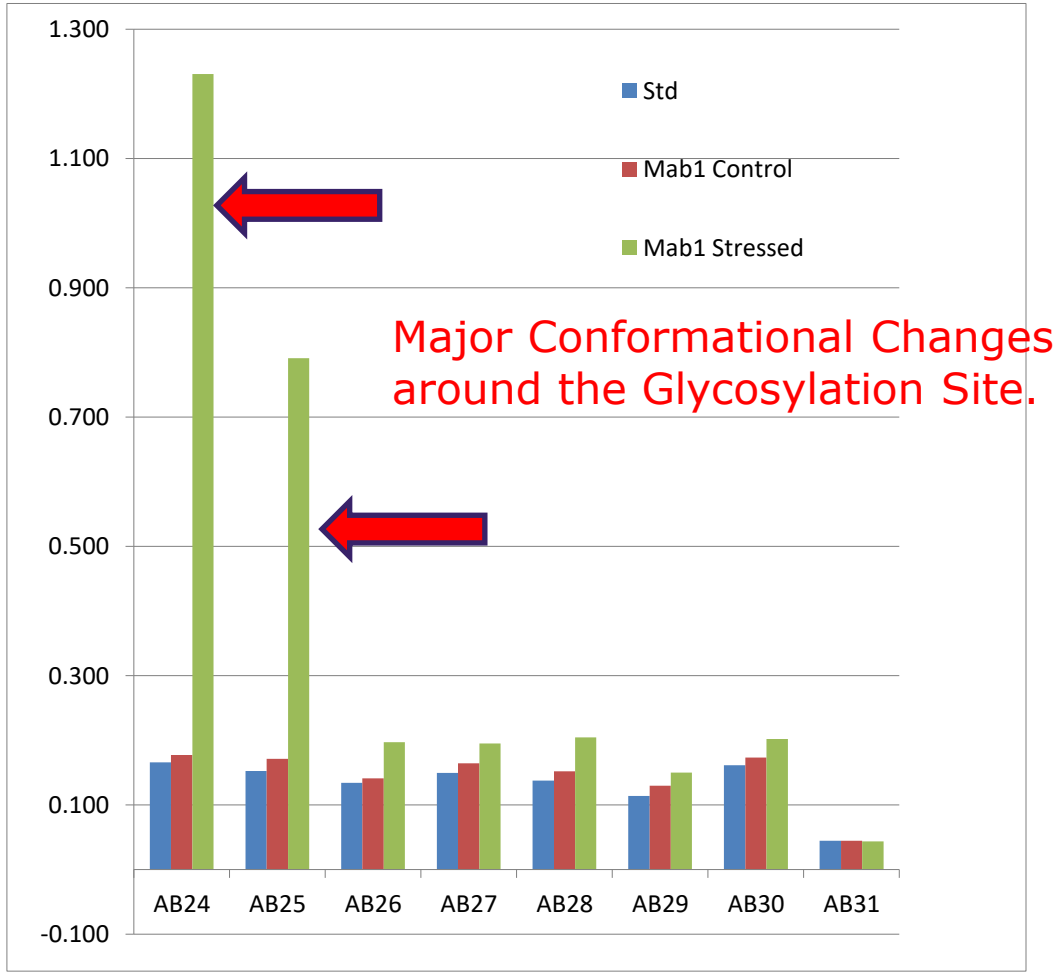


Ab6 is close to light chain CDR3 22% Bioassay Activity Decrease

Case study: Correlation Between Conformation and Bioassay in Stability Testing (Novel mAb) continued.

FcγRIIIa binding result: 64% Decrease

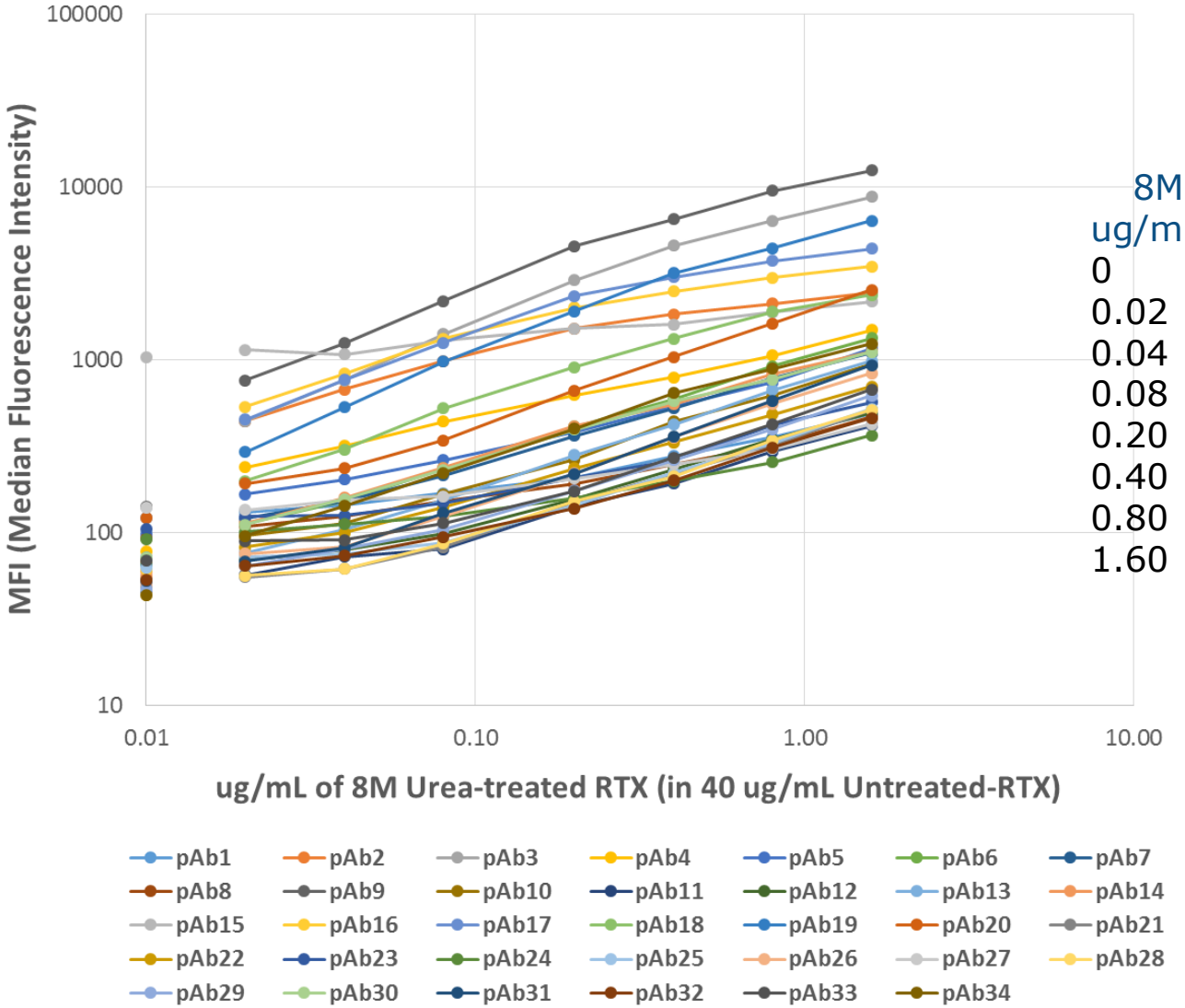




Ab15,16: LC Hinge Region; Ab17,18:HC, Fv-Fc domain
Ab24: HC Hinge Region.; Ab25: HC Glycosylation Site.

Sensitivity: Spiking with 8M Urea-treated mAb

As low as 0.05% epitope exposure can be detected and quantified



8M Urea-RTX	
ug/mL	% Spiked
0	0
0.02	0.05%
0.04	0.10%
0.08	0.20%
0.20	0.50%
0.40	1%
0.80	2%
1.60	4%

Currently available assays.

InnoBridge is designed for novel mAb Development.

mAb Name	Trade Name	Composition	IgG Class	Sales ($ billions)
Bevacizumab	Avastin	Humanized mAb	IgG1	7.0
Cetuximab	Erbitux	Humanized mAb	IgG1	2.3
Alemtuzumab	Campath	Humanized mAb	IgG1	0.7
Rituximab	Rituxan	Chimeric mAb	IgG1	8.6
Adalimumab	Humira	Human mAb	IgG1	16.1
Trastuzumab	Herceptin	Humanized mAb	IgG1	6.9
Palivizumab	Synagis	Humanized mAb	IgG1	0.5
Infliximab	Remicade	Chimeric mAb	IgG1	10.2
Etanercept	Enbrel	Fc Fusion Protein	IgG Fusion	9.1
Erythropoietin	EPO	Human protein	Non-mAb	3.4
Pegfilgrastim	Neulasta	Human protein	Non-mAb	4.6
Denosumab	Prolia	Human mAb	IgG2	3.0
Ranibizumab	Lucentis	Humanized mAb	IgG1 Fab	4.3
Golimumab	Simponi	Human mAb	IgG1	2.9
Ustekinumab	Stelara	Human mAb	IgG1	2.5
Aflibercept	Eylea	VEGFR-1-Fc Fusion Protein	IgG Fusion	5.9
Somatropin	Genotropin etc.	Human Growth Hormone	Non-mAb	5.2

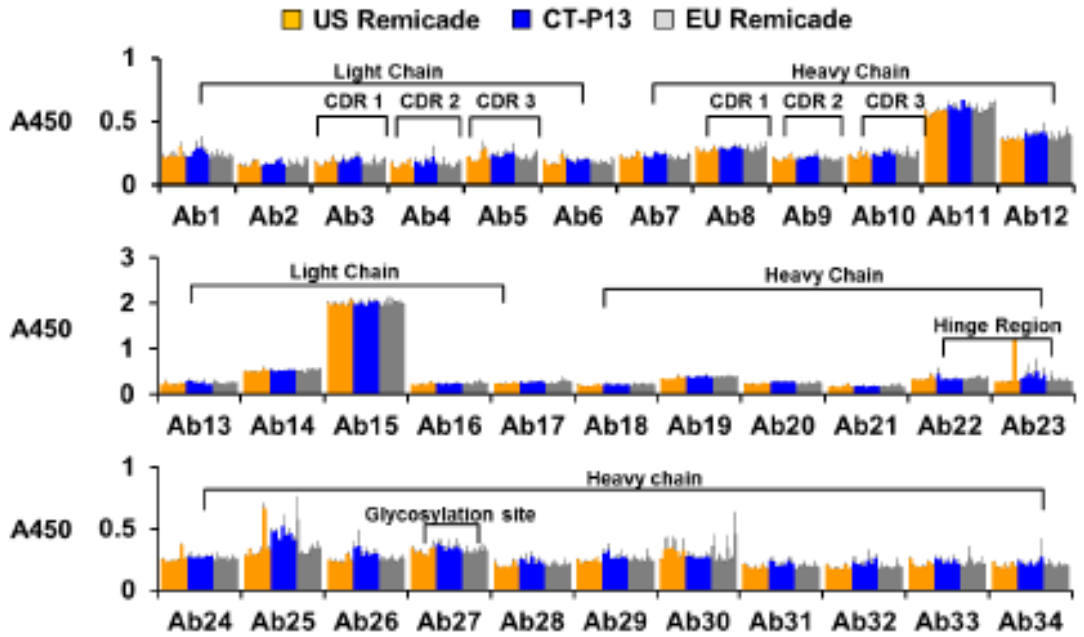
CT-P13 Infliximab BioSimilar*: PCA used in an FDA Submission.

8.4 IMMUNOGENICITY RESULTS

8.4.1 Antibody Array Study

Antibody array technology or Protein Conformational Array (PCA) is a technique for comparing structural differences among similar molecules such as monoclonal Antibodies (mAb). The ELISA consists of a pool of 34 pAbs, each raised against a short segment of the linear mAb peptide sequence. Together, this overlapping series of peptides covers the entire peptide sequence of the mAb and were the mAb to exist in a linear or denatured state, each of the 34 pAbs would give a strong signal in the ELISA. With a correctly folded mAb, most of the epitopes are buried and are not strongly recognized by the pAbs. Difference in intensity of the responses for each pAb reflects the exposure of the epitope Ab detects. This technique showed CT-P13, EU Remicade, and US Remicade were consistent with regard to epitope exposure and higher order structure (Figure 54). One batch of US Remicade (CJM76016P1) showed deviations at some epitopes around the hinge region and in the overlapping region of 254-275 aa and 272-293 aa of the HC, suggesting slight unfolding in this region.

ure 54: Antibody Array Data Showing Epitopes Exposed by 7 Lots each of US Remicade, CT-P13, and EU Remicade using 34 Polyclonal Sera



CDR: Complementarity determining region

- Notes differences between US & EU Remicade production and CT-P13.

HOS similarity between CT-P13 (Biosimilar) and its originator manufactured in the EU & USA.

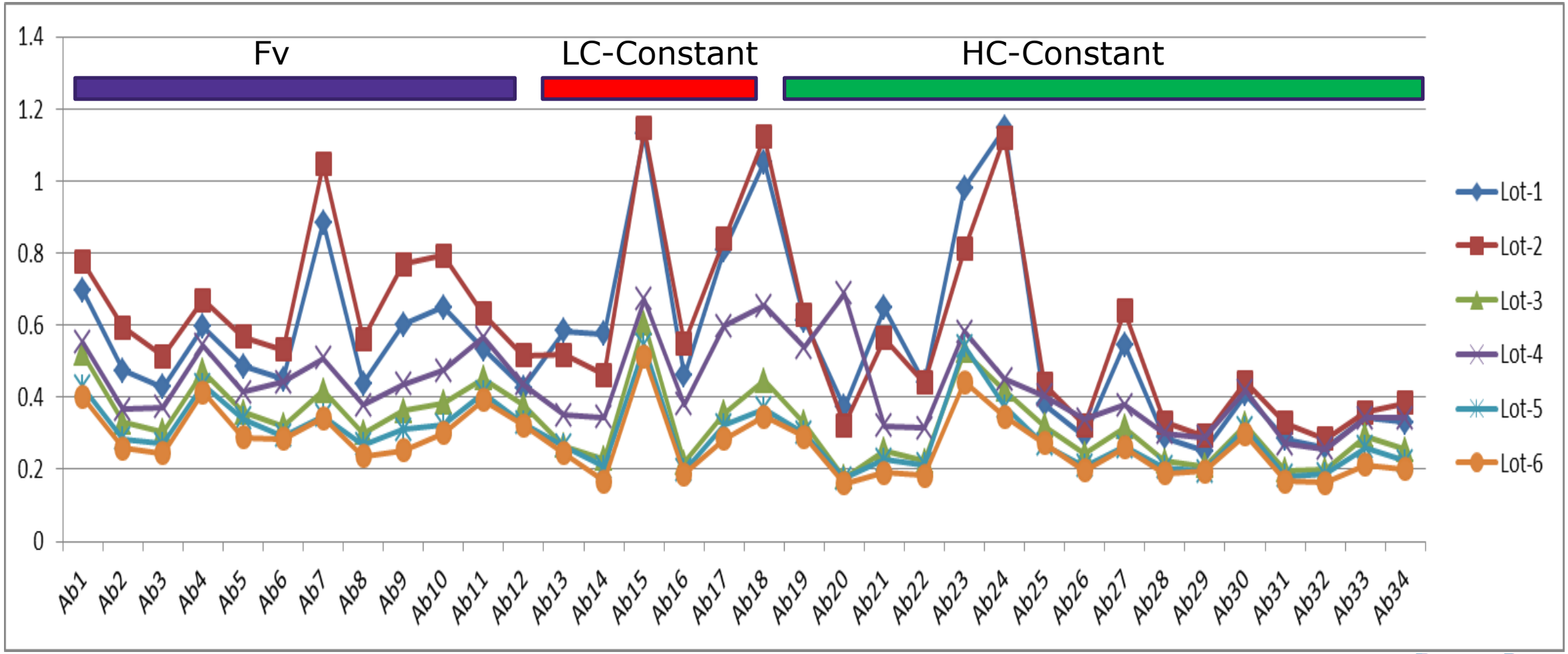
***CT-P13 (infliximab biosimilar) BRIEFING DOCUMENT FOR THE ARTHRITIS ADVISORY COMMITTEE, MEETING DATE: February 9, 2016.**

FDA Advisory Committee Briefing Document, February 9, 2016, Page **1** of **349 (pg179)**

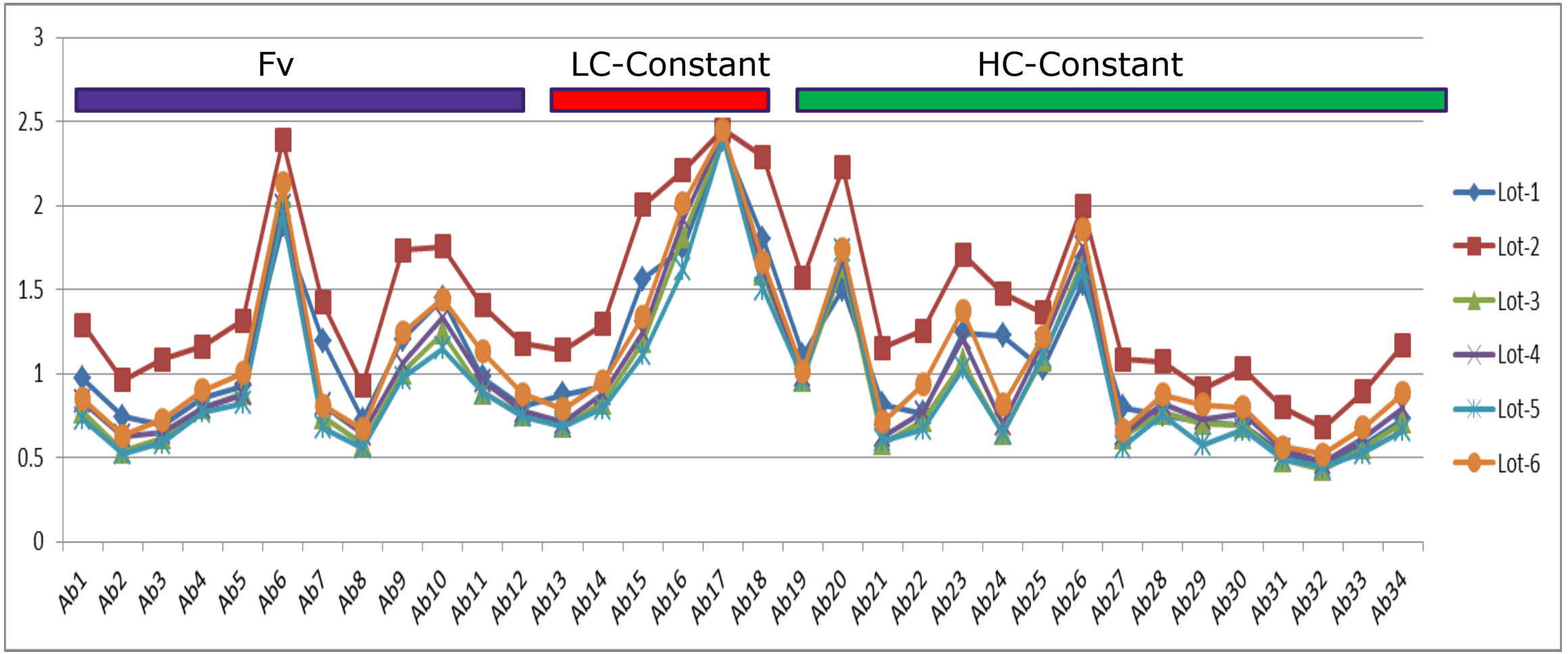
Potential Applications for PCA Technology

- Biosimilar as well as Novel mAb Development
- Cell Line Selection
- Process Development
- Formulation Development
- Comparability Studies
- Product Characterization
- An Easy and Accurate ID Test
- Antibody-Drug Conjugates (ADCs)

Herceptin without Heat Treatment (55°C Overnight)

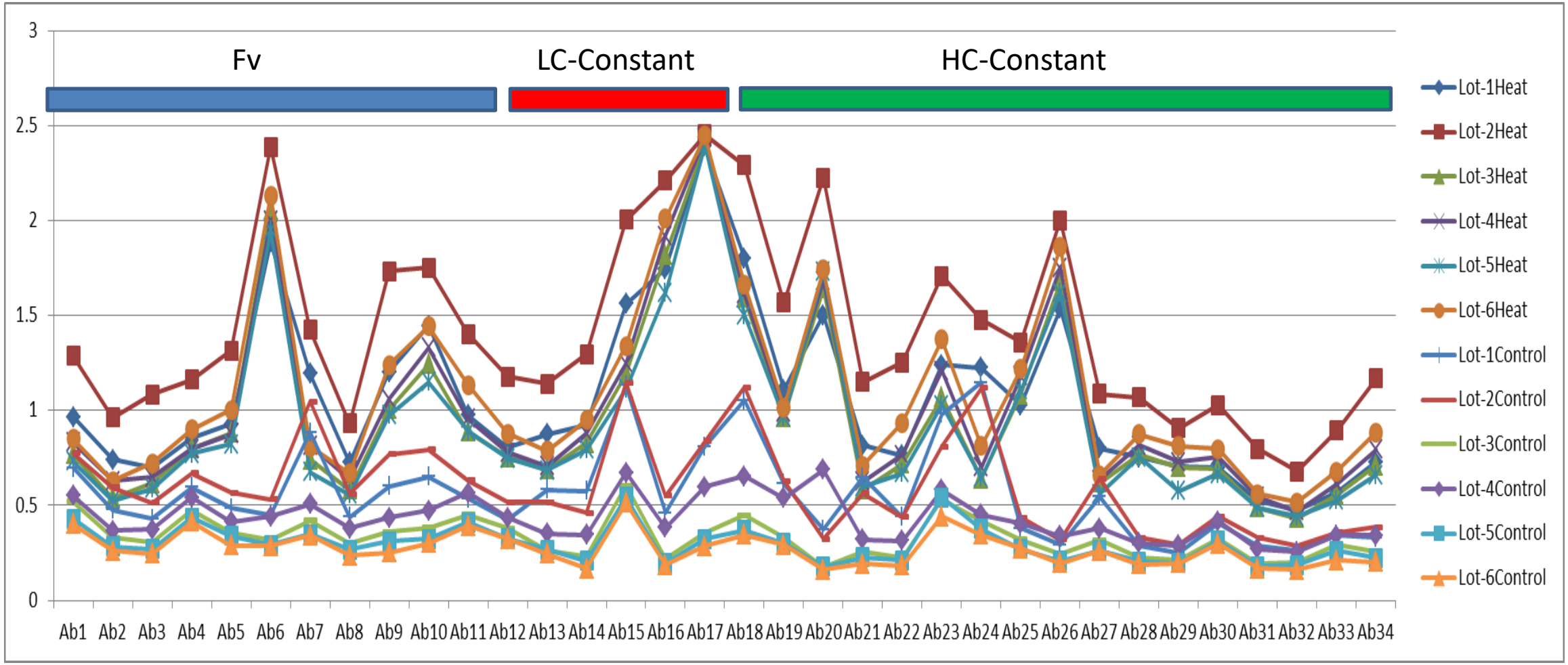


Herceptin with Heat Treatment (55°C Overnight)



34-PCA Antibody Panel

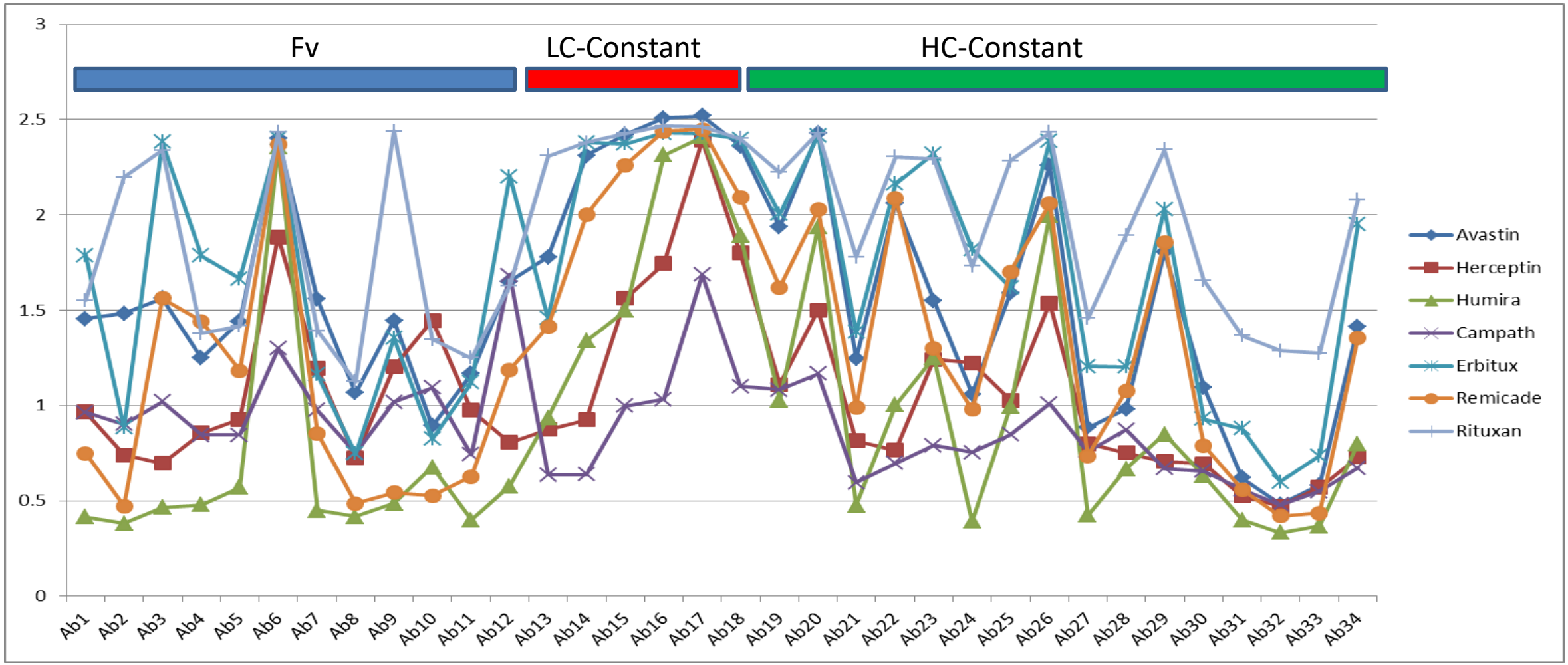
Herceptin with and without Heat Treatment



34-PCA Antibody Panel

HOS Stability Profiles of 7 Marketed mAbs

Each mAb has a unique HOS profile

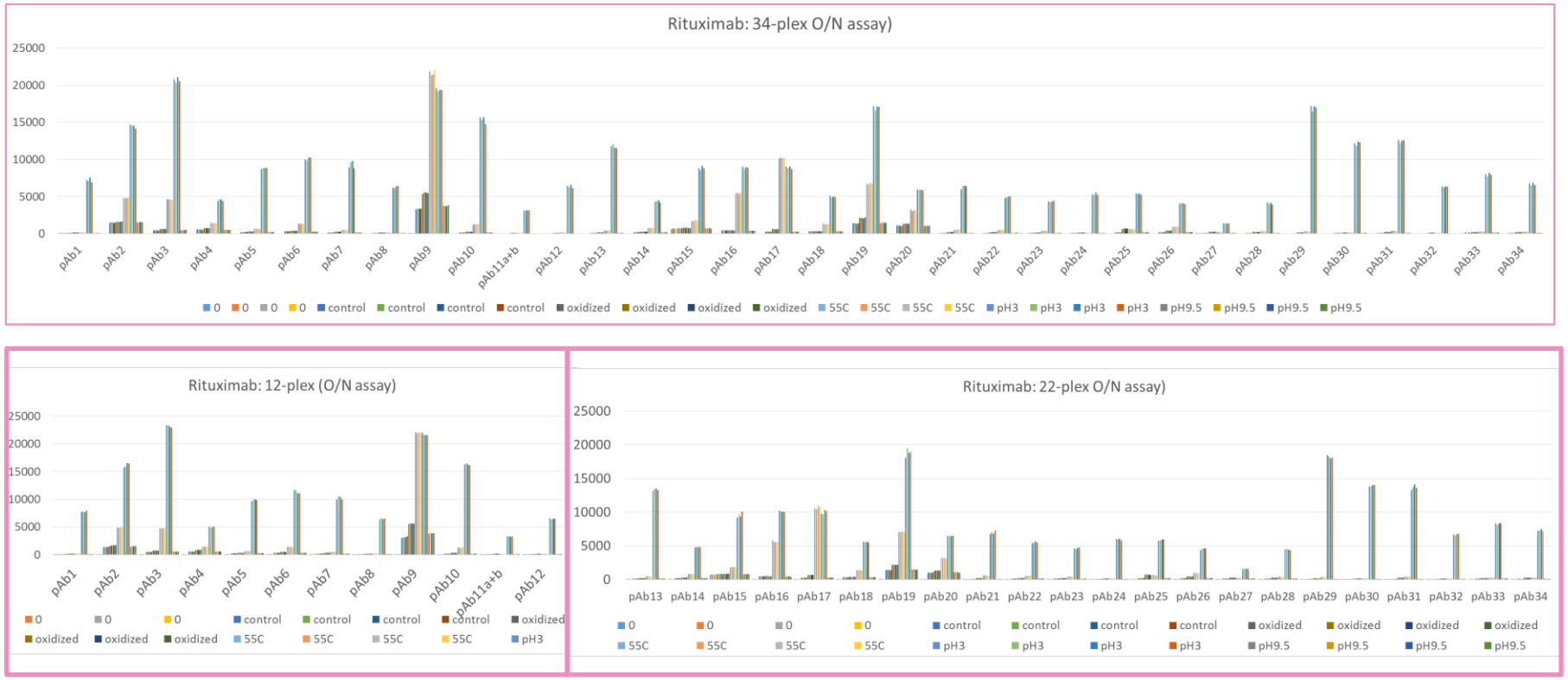


34-PCA Antibody Panel

3. Developing PCA : from ELISA to xMAP.

RituBridge to RituPlex Example

Multiplex: a 34-plex = a 12-plex + a 22-plex

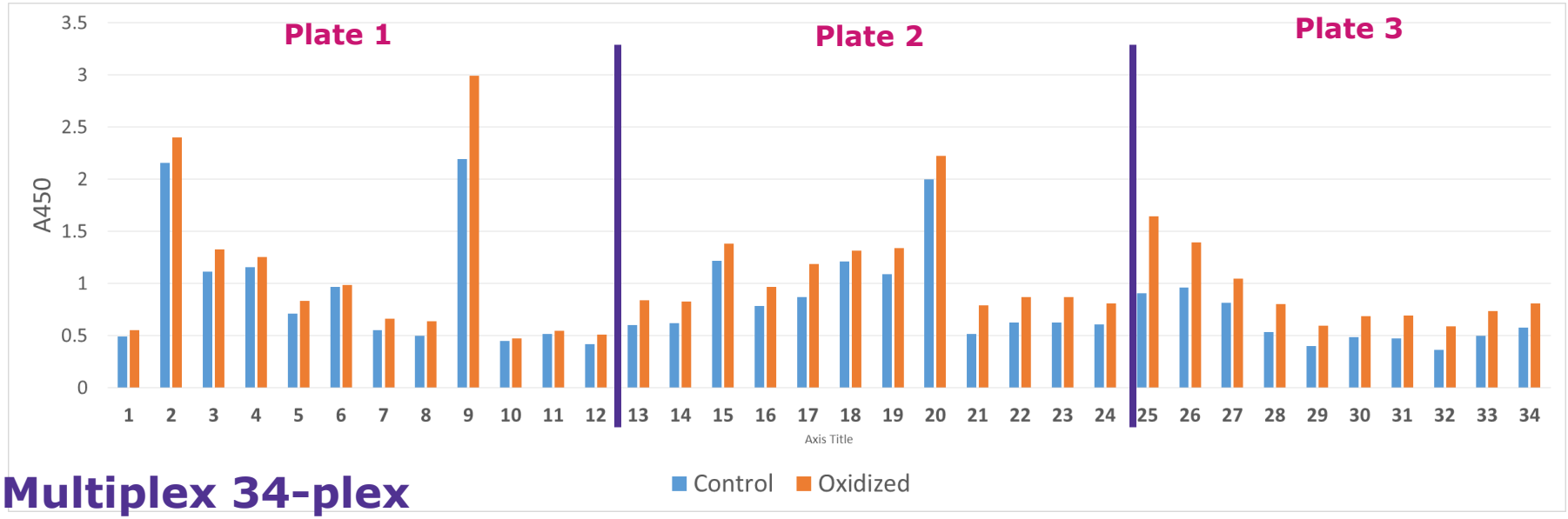


Variable Regions Constant Regions

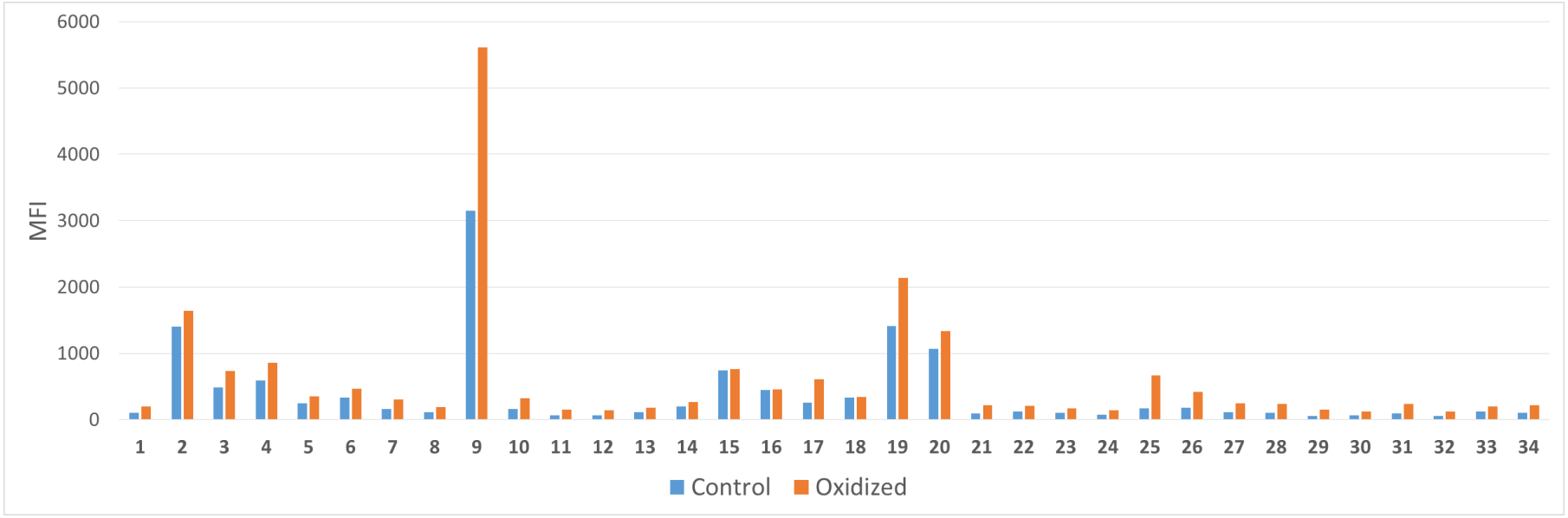
- **Multiplex can be formatted as either a 34-plex or a 12-plex + a 22-plex**
- MFI (Median Fluorescence Intensity) data from background (no sample), control (not-treated rituximab), oxidized, 55C, pH3, pH9.5 -treated rituximab

ELISA vs. Multiplex: Control vs. Oxidized Rituximab

ELISA's



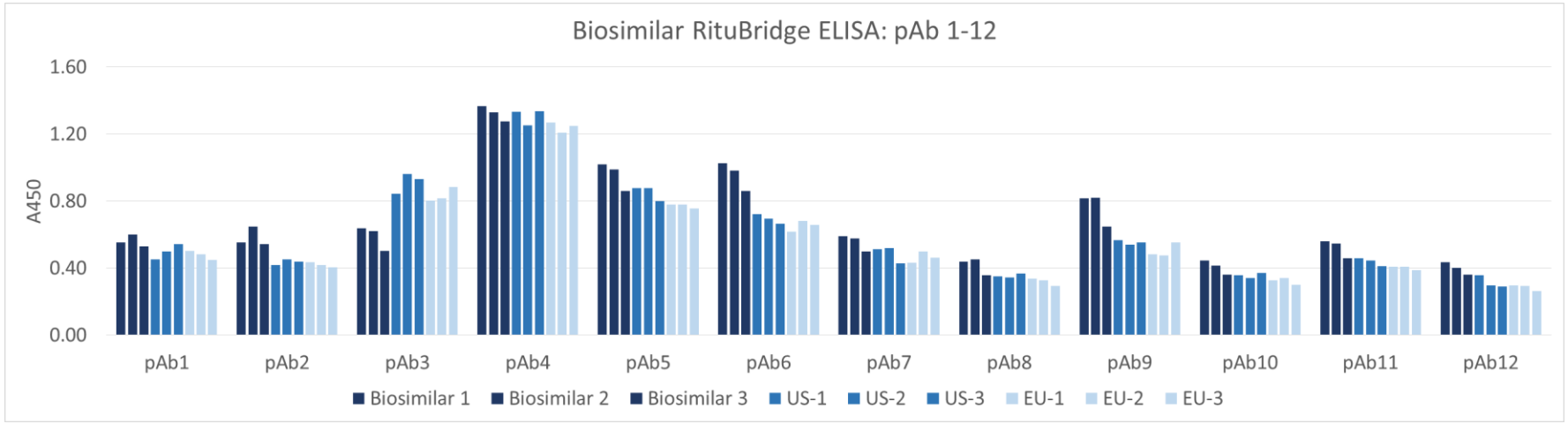
Multiplex 34-plex



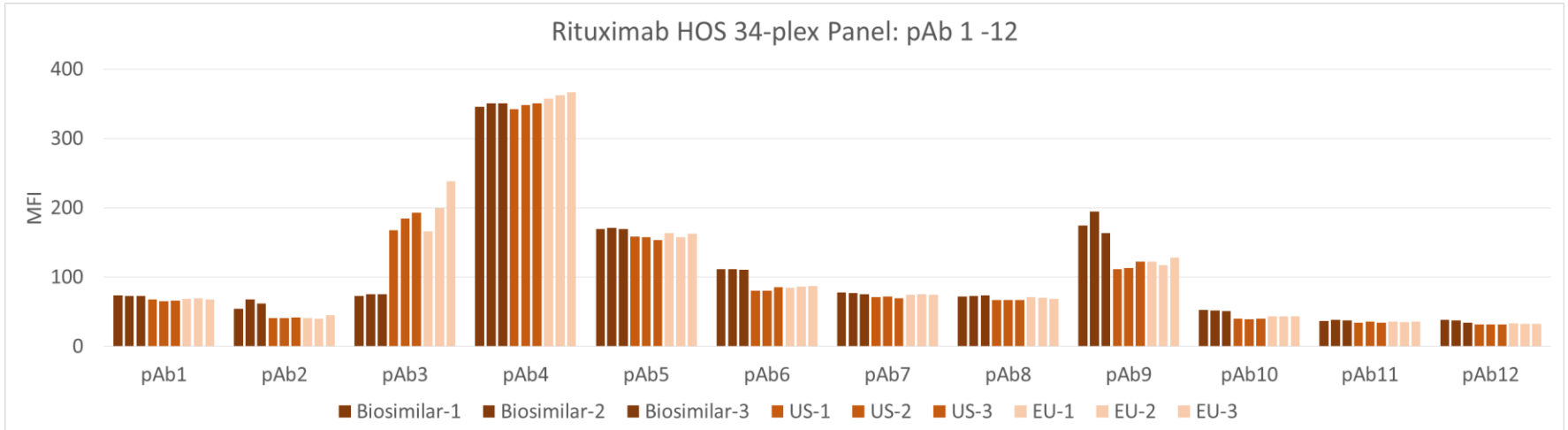
ELISA to Multiplex a Comparison 2: Rituximab Reference vs. Biosimilar.

ELISA vs. Multiplex: pAb 1-12 (Variable Regions)

ELISA Plate-1

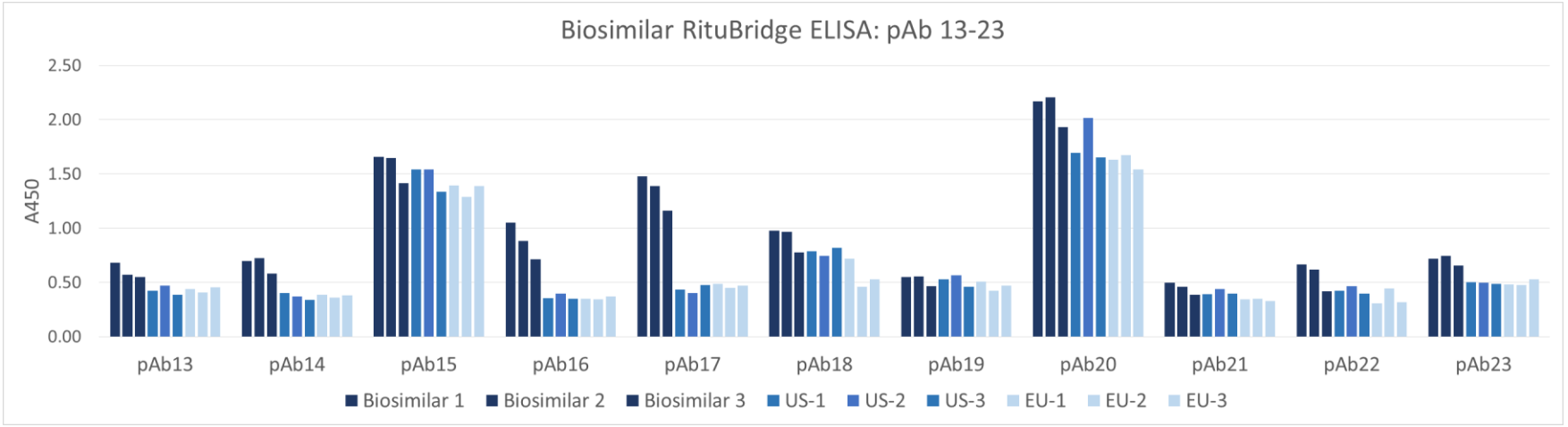


Multiplex

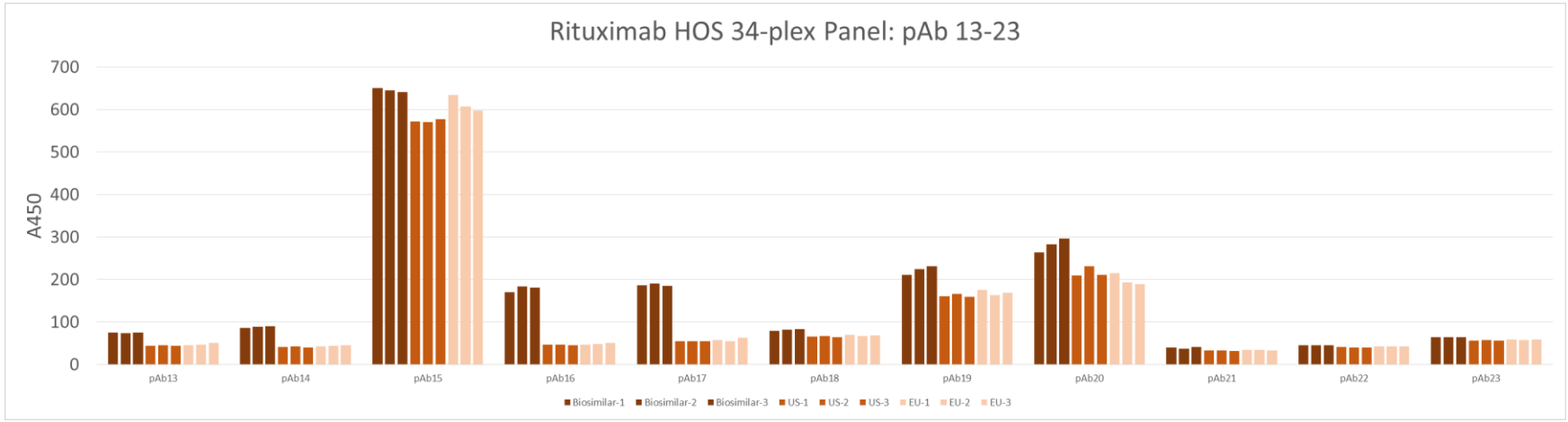


ELISA to Multiplex: pAb 13-23 (Constant Regions-1)

ELISA Plate-2

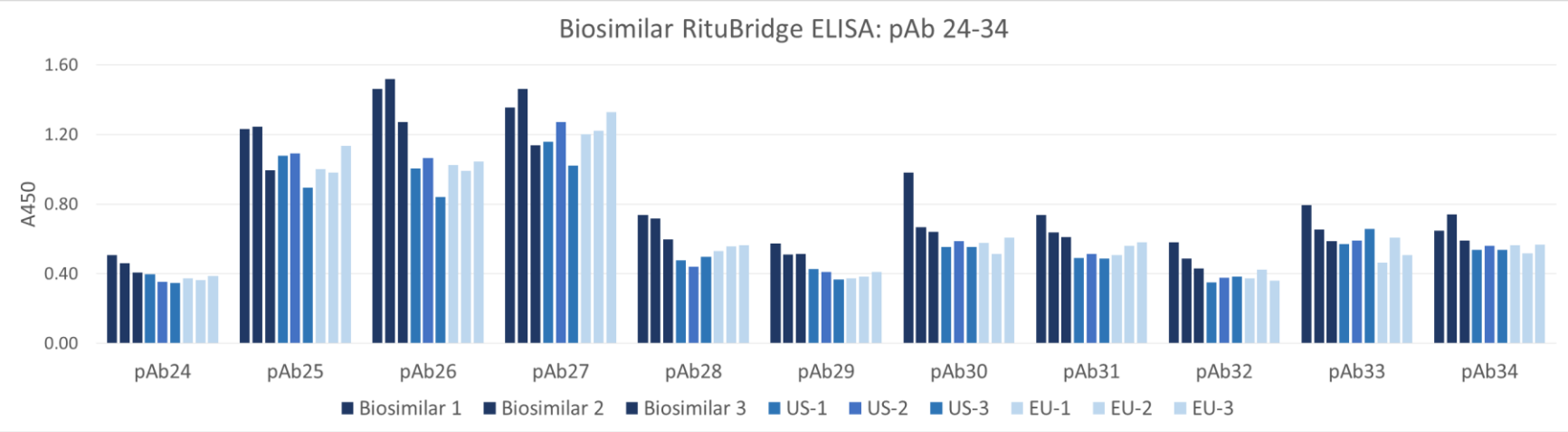


Multiplex

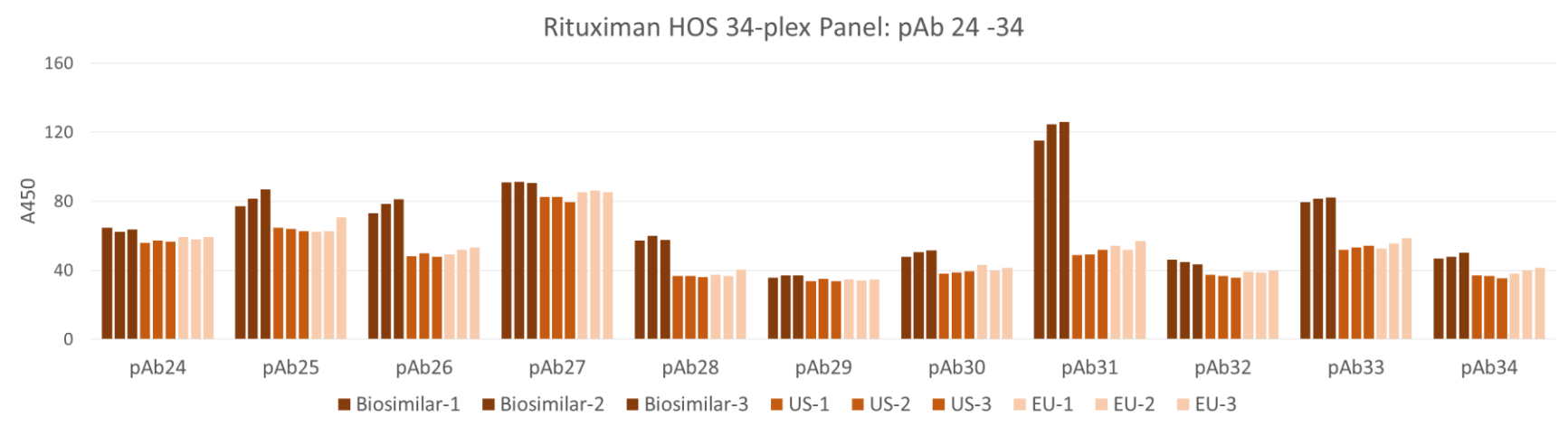


ELISA to Multiplex: pAb 24-34 (Constant Regions-2)

ELISA Plate-3



Multiplex



INNOPLEX: Publication*

MABS
2018, VOL. 0, NO. 0, 1–9
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REPORT

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Monoclonal antibody higher order structure analysis by high throughput protein conformational array

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Bristol-Myers Squibb

ABSTRACT

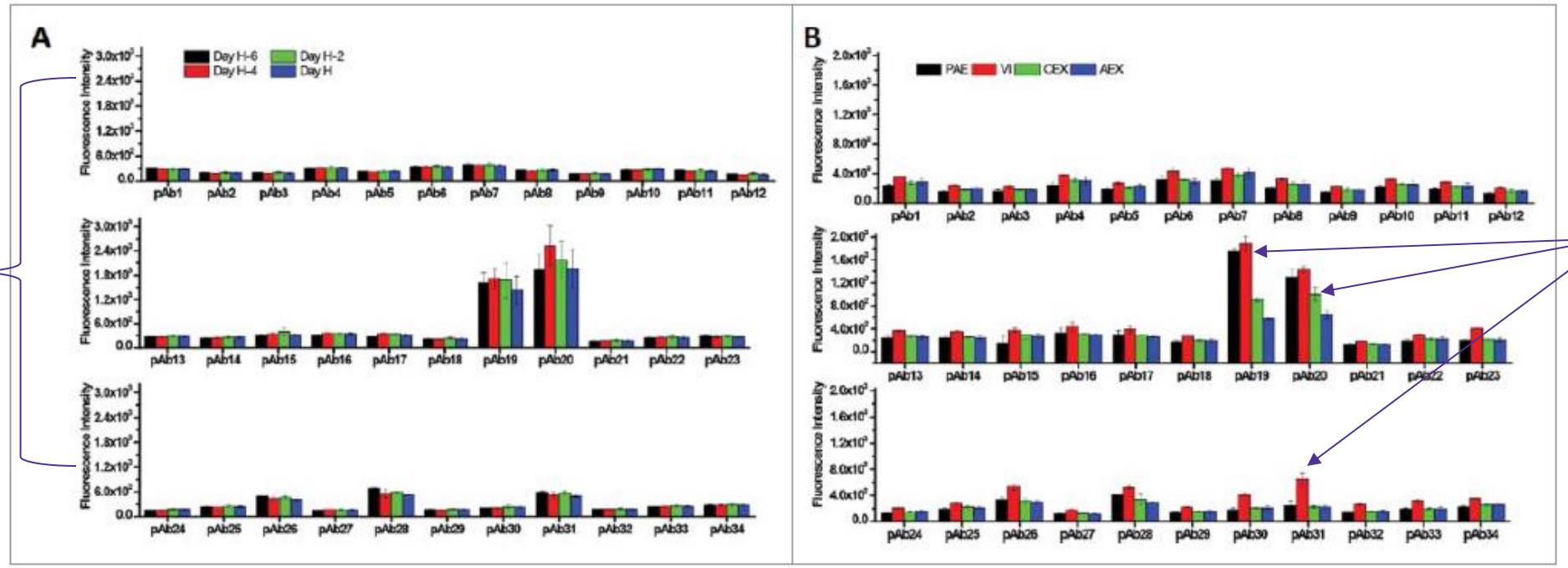
The elucidation of antibody higher order structure (HOS) is critical in therapeutic antibody development. Since HOS determines the protein bioactivity and chemo-physical properties, this knowledge can help to ensure that the safety and efficacy attributes are not compromised. Protein conformational array (PCA) is a novel method for determining the HOS of monoclonal antibodies. Previously, we successfully utilized an enzymelinked immunosorbent assay (ELISA)-based PCA along with other bioanalytical tools to elucidate the structures of antibody aggregates. In this study, applying **a new multiplex-based PCA with 48-fold higher throughput** than the ELISA-based one **we revealed structural differences between different antibody molecules and antibody structure changes affected by various processing conditions**. The PCA analysis of antibody molecules clearly demonstrated significant differences between IgG1 and IgG4 subclasses in epitope exposure and folding status. Furthermore**, we applied small angle X-ray scattering to decipher mechanistic insights of PCA technology and validate structural information obtained using PCA**. These findings enhance our fundamental understanding of mAbs' HOS in general. **The PCA analysis of antibody samples from various processing conditions also revealed that antibody aggregation caused significantly higher exposure of antibody epitopes, which potentially led to a "foreign" molecule that could cause immunogenicity**. **The PCA data correlated well with protein stability results from traditional methods such as size-exclusion chromatography and protein thermal shift assay**. Our study demonstrated that high throughput PCA is a suitable method for HOS analysis in the discovery and development of therapeutic antibodies.

*Monoclonal antibody higher order structure analysis by high throughput protein conformational array. Y.Song *et al*, MABS 2018 Jan 9, 1-9.

Process applications highlighted within MABS publication.

Cell culture

Seems stable



Process driven changes

Figure 14: **(Fig 5)**(A) PCA data of mAb5 samples from upstream process development. Samples were collected during the cell culture on Day H-6, H-4, H-2, and H as labeled (Day H is the harvest day). The error bar is the standard deviation from two repeats. (B) PCA data of mAb5 samples from downstream process. Samples collected include ProteinA Elution (PAE), Virus Inactivation (VI), Cation Exchange Elution (CEX), and Anion Exchange Flow-through (AEX). The error bar is the standard deviation from two repeats.

"we revealed structural differences between different antibody molecules and antibody structure changes affected by various processing conditions"

ELISA to xMAP: Summary

- We developed a multiplex panel for HOS analysis of rituximab biosimilar. All 34 pAb can be run simultaneously in a single well.
- The multiplex vs. ELISA side-by-side studies demonstrated a similar data profile.
- 8 M urea treated mAb (unfolded mAb so that linear epitopes become easily accessible) can be used to estimate the extent of epitope exposure in native samples.
- The multiplex assay has the advantages over ELISA on very little sample, little hands-on time, very fast, high precision, and wider dynamic range.

2-3 Samples per Kit.

3 plates with in the product create the array

40+Samples per Kit.

Each well contains the full array.

The assays original ELISA format allows for a low barrier to the technologies utilization. Higher volumes of samples produced through the process development phases dictate a move to a higher throughput format is required for which we will use the Luminex® xMAP platform.

4. The Immunogenicity and Higher Order Structure Correlation Study

Studies Demonstrating the Importance of 3-D Structure and Its Stability for Immunogenicity

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Consequences of anti-drug antibodies

Loss of efficacy
- Insulin
- Streptokinase
- Staphylokinase
- ADA
- Calcitonin
- Factor VIII
- Interferon alfa 2
- Interferon beta
- Interleukin-2
- GnRH
- TNFR55/IgG1
- Denileukin diftitox
- HCG
- GM-CSF/IL3
- Various monoclonals

Enhancement of efficacy
- Growth hormone

Neutralization of endogenous protein
- Epoetin
- Megakaryocyte-derived growth factor (MDGF)

General immune effects
- Allergy
- Anaphylaxis
- Serum sickness, etc

None

From Wim Jiskoot, NBC, Seattle, 2009

Cytokine Release Experimental Design

1. 3 antibodies used: IgGs from human plasma; Herceptin, Rituxan.

2. 6 different conditions tested: no treatment; 8 M urea-induced unfolding; Heat treatment; Oxidation; Higher pH (pH 9.5); Lower pH (pH3).

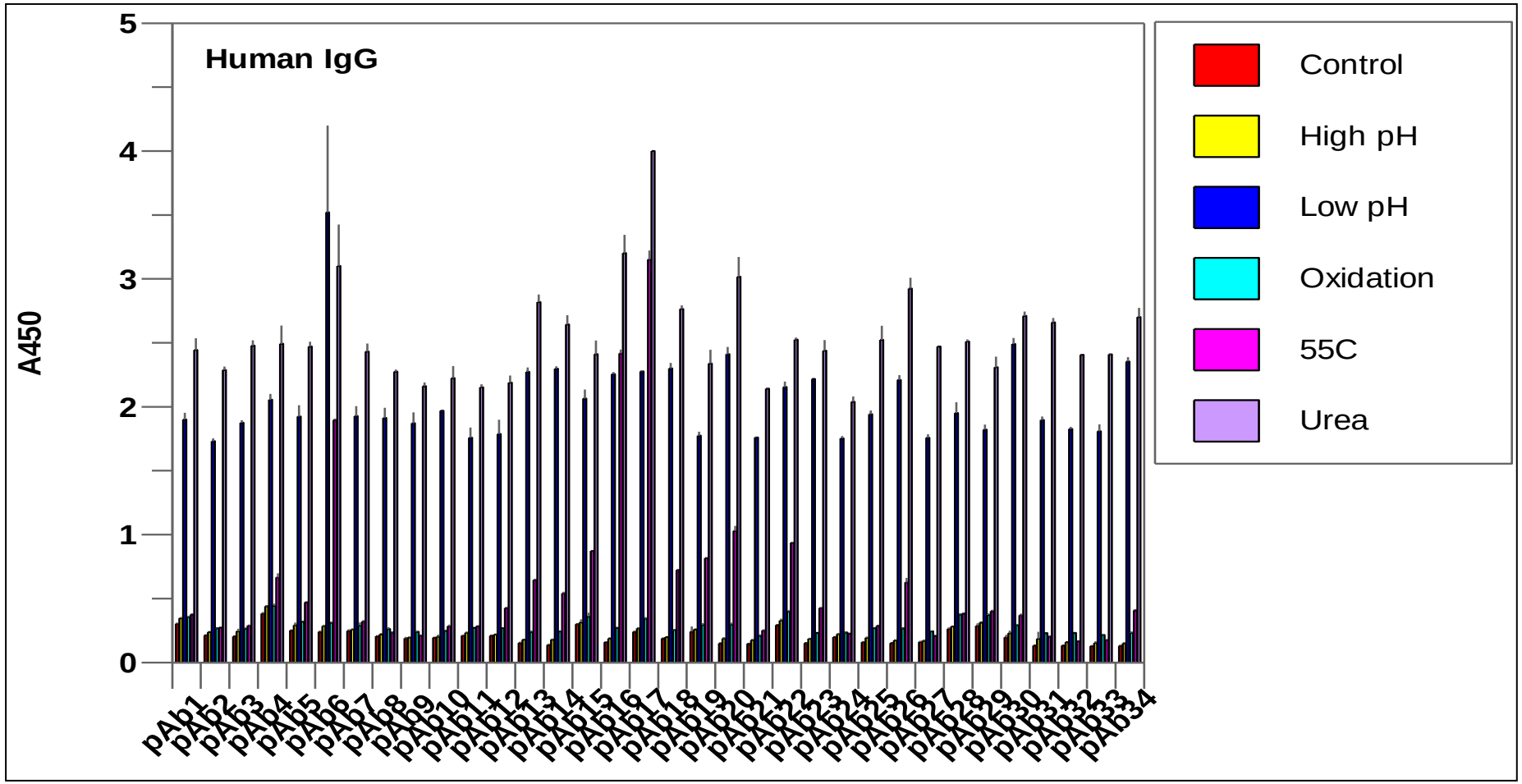
3. Three antibody levels tested: 100 µg/mL; 10 µg/mL; 1 µg/mL (mimic the levels in the human blood during actual treatment).

4. Positive and negative controls: PBS will be used as the negative control to determine assay background and base-line cytokine levels, LPS stimulation will be used as a positive control for measuring cytokine release. Urea (screen at 3 different concentrations) will be tested as the vehicle control for urea-induced unfolding.

5. Plasma samples from the whole blood incubation are analyzed with HSTCMAG384-PX21 High Sensitivity Human Cytokine Assay kit from MilliporeSigma (measuring 21 cytokines simultaneously) on a Luminex FlexMap3D Analyzer.

6. Human Whole Blood Cytokine Release Test (n=5) (Preliminary analysis of 11 selective cytokines from the 21 cytokines measured)

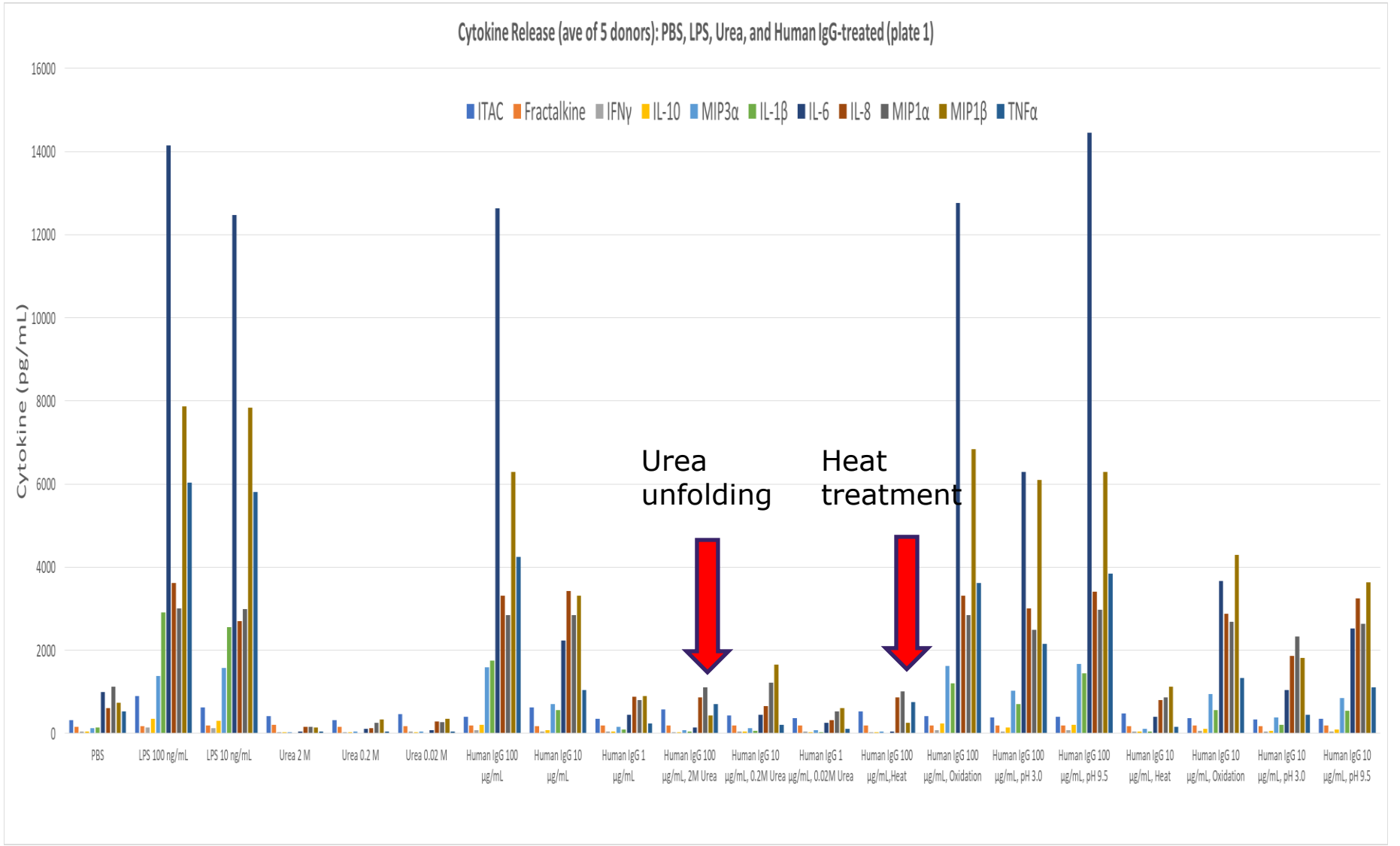
HOS Changes from Serum-derived Human IgGs Under Different Stress Conditions



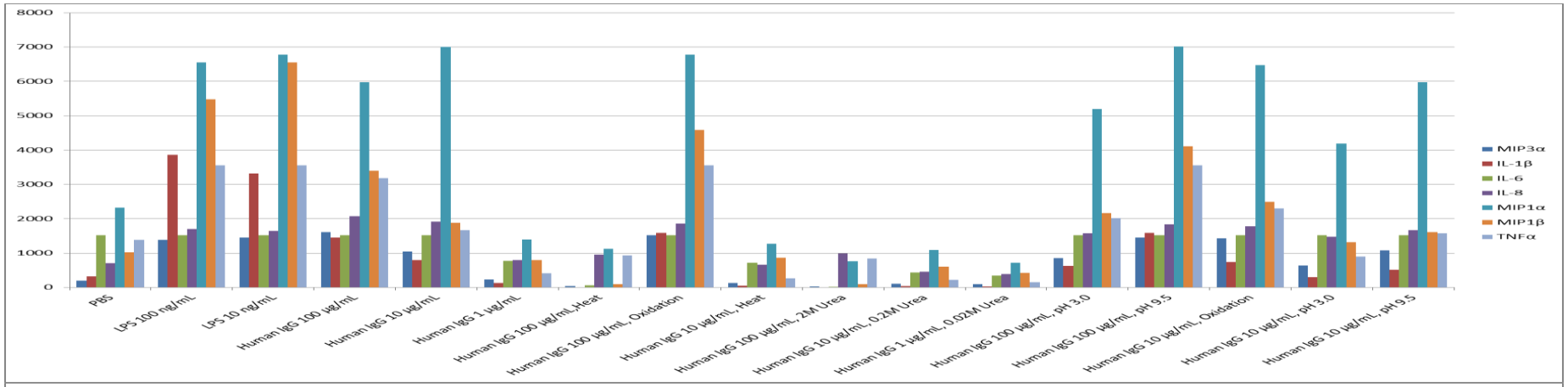
The HOS of Human serum-derived IgGs are stable in high pH (pH9.5) and oxidation, Changes at different region with low pH (pH 3.0) and heat treatment (55°C, O/N).

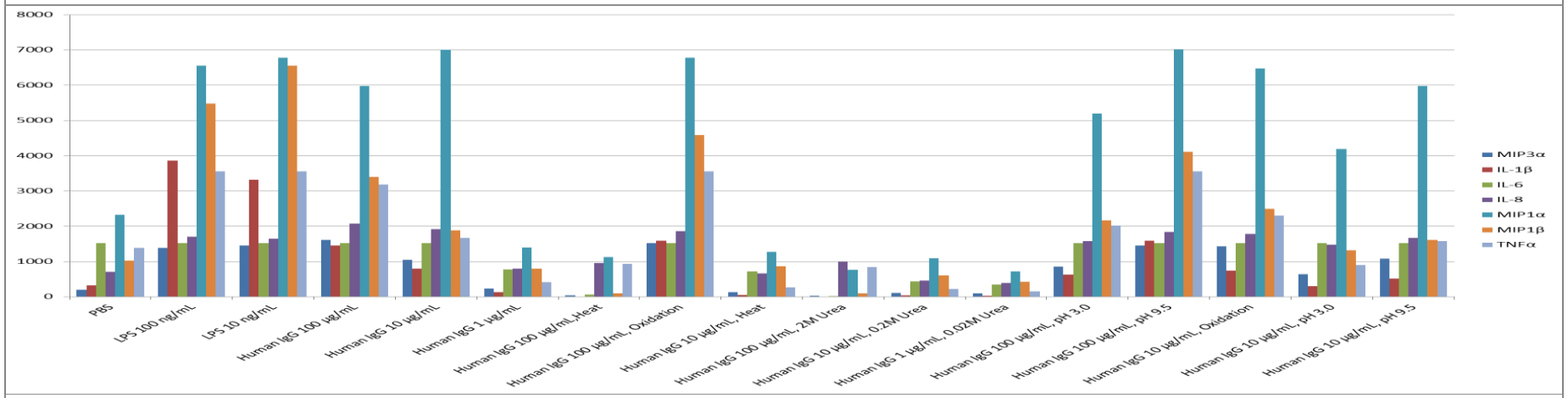
There is general unfolding in 8 M urea but refolding is fast.

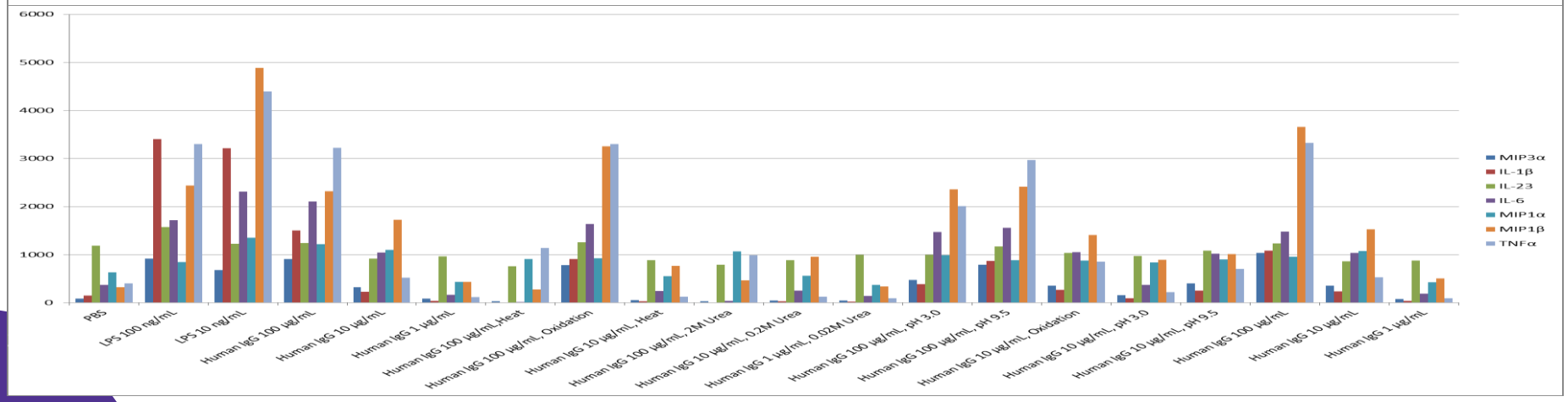
Human Whole Blood Cytokine Release with IgGs Purified from Human Serum



NB: Only 11 of the 21 cytokines measured was shown here.



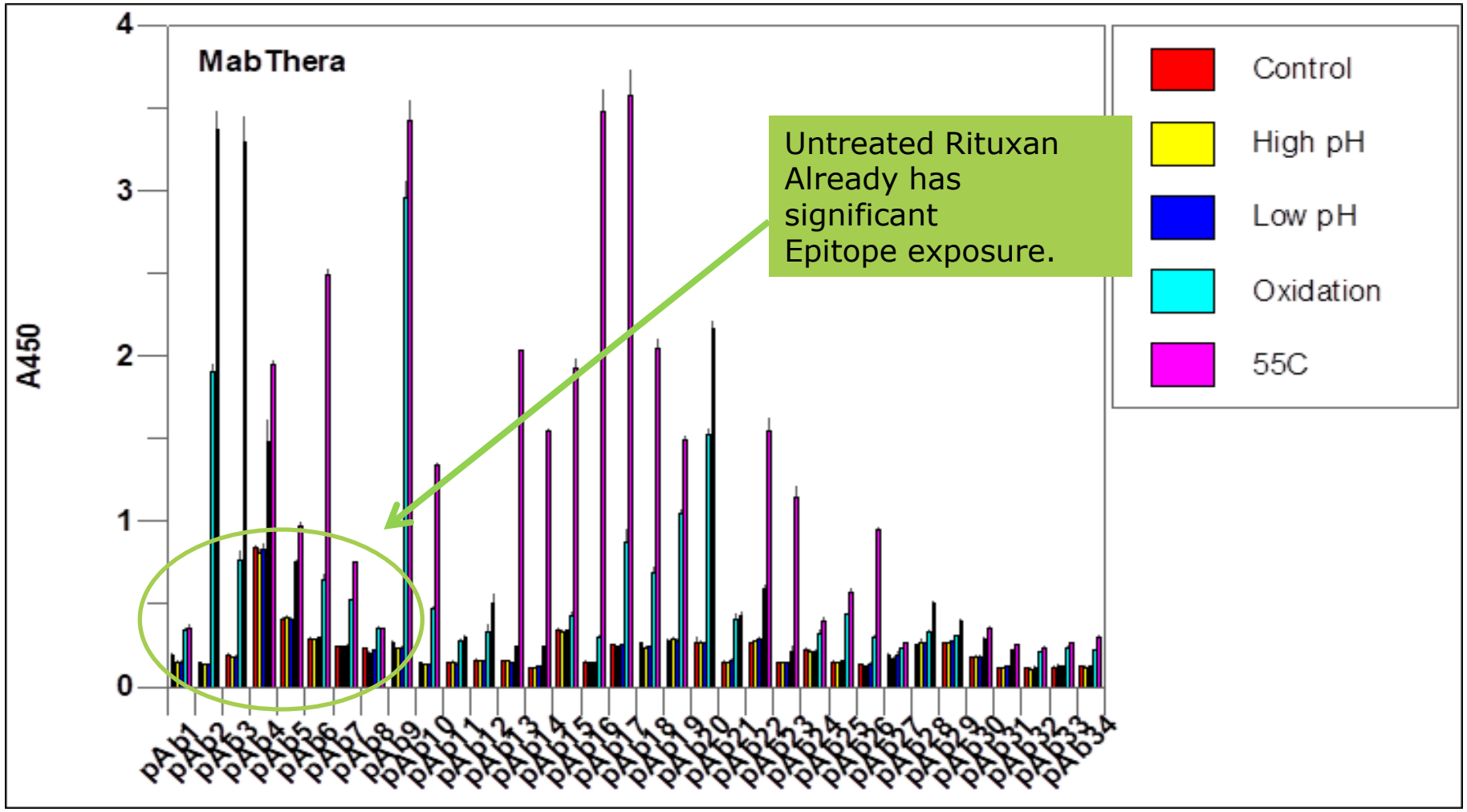




Donors 1, 2 and 3:

Responses to human IgGs differently.

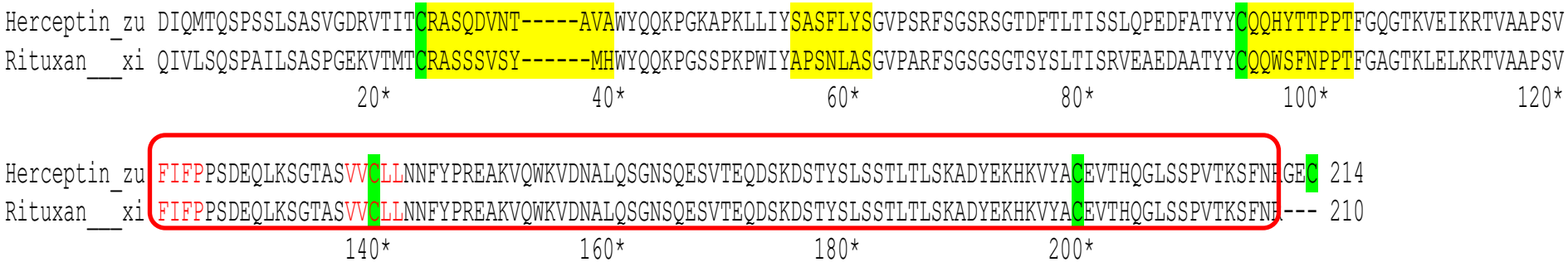
HOS Changes from Rituxan Under Different Stress Conditions



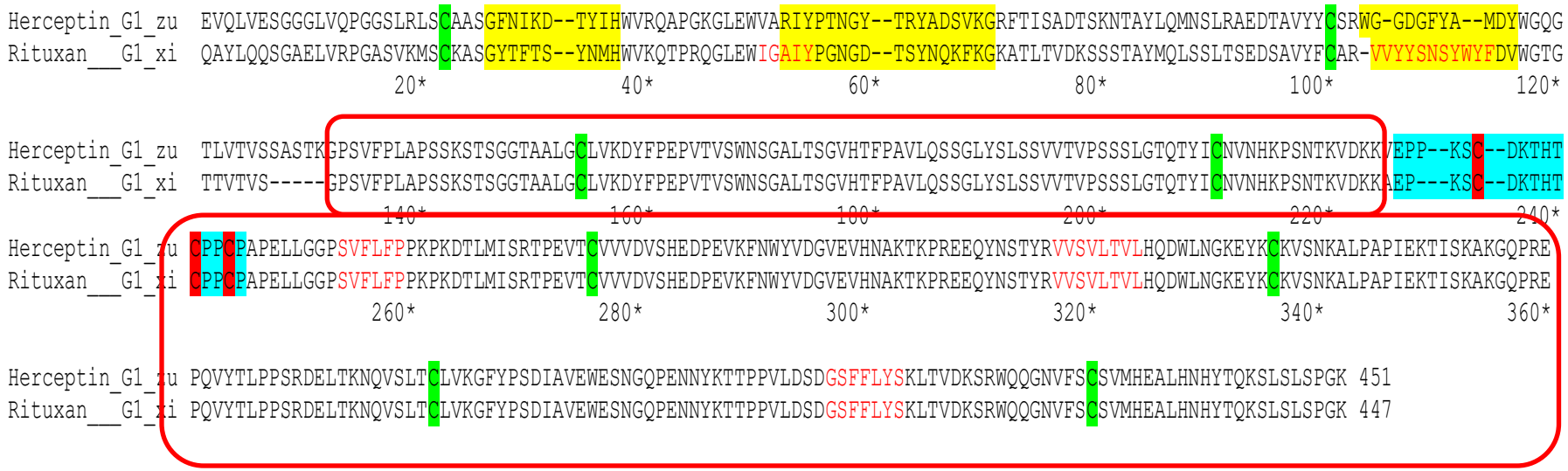
The HOS Changes at different region with high and low pH (pH 9.5 and 3.0 respectively), oxidation and heat treatment (55°C, O/N). There is general unfolding in 8 M urea and refolding is slow.

Herceptin and Rituxan Light Chain Comparison

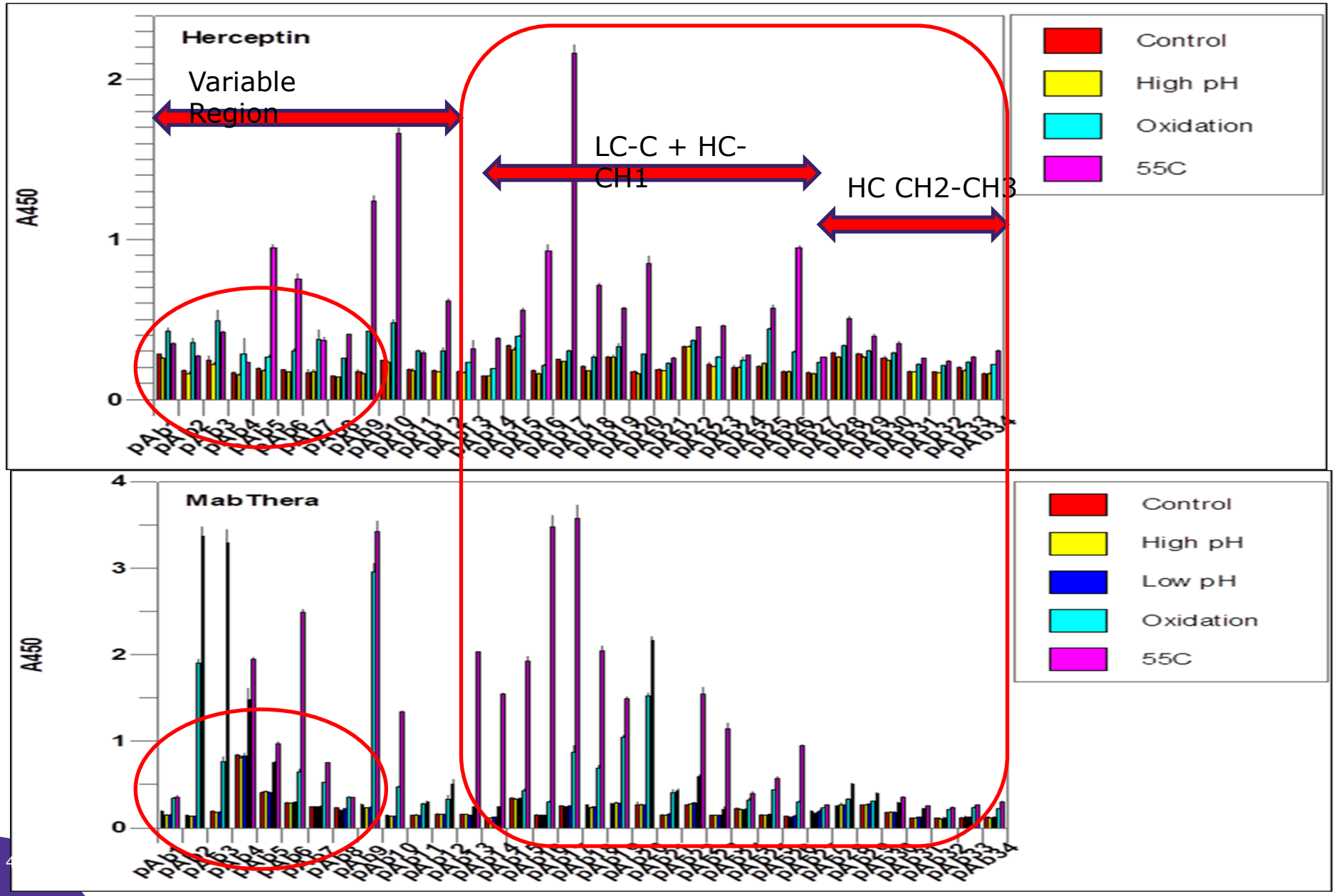
Constant regions are identical.



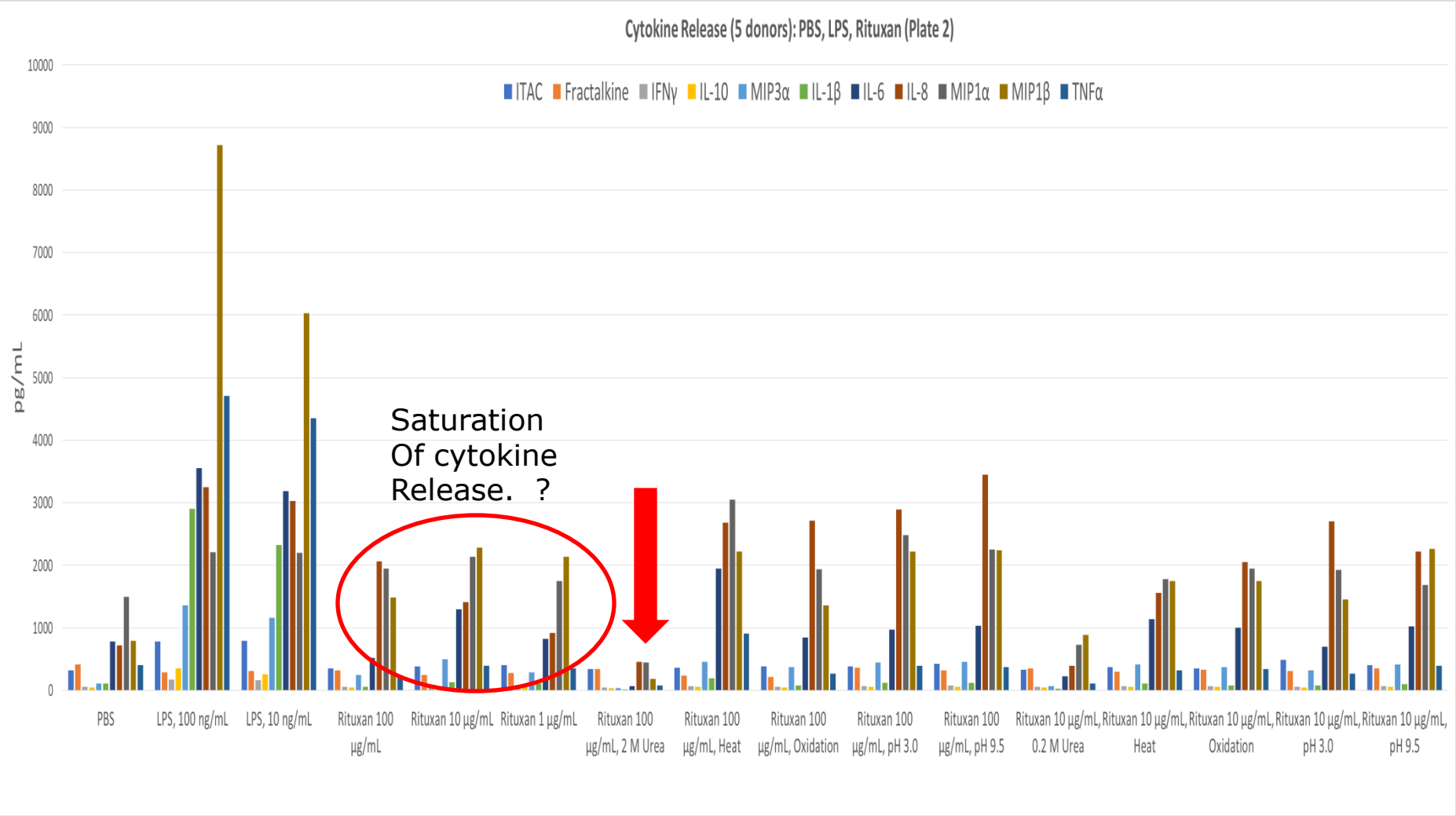
Herceptin and Rituxan Heavy Chain Comparison



Comparison of Herceptin and Rituxan HOS Stability (Different Scale)
Herceptin seems more stable than Rituxan under stress conditions



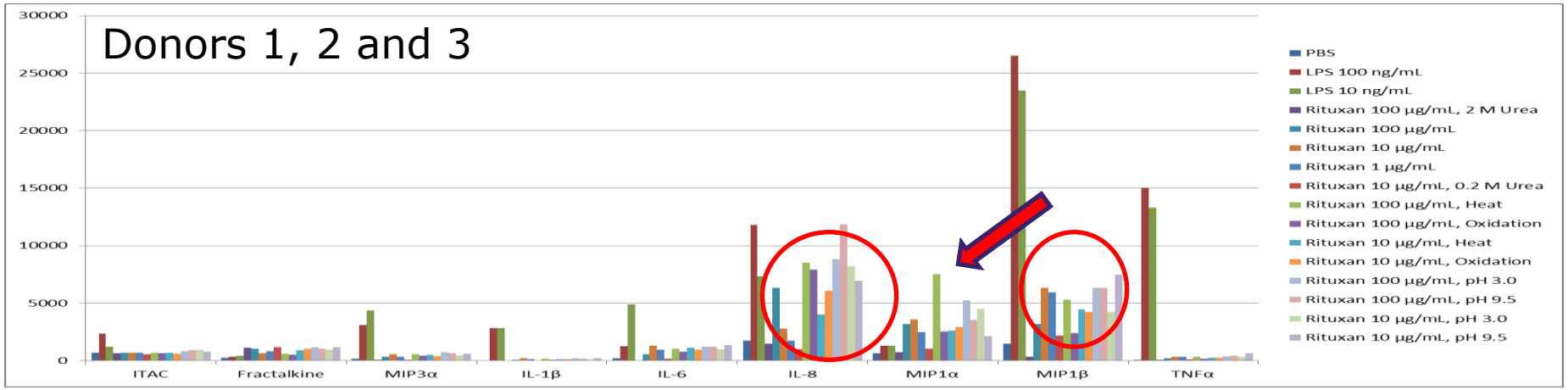
Human Whole Blood Cytokine Release with Rituxan Under Different Stress Conditions

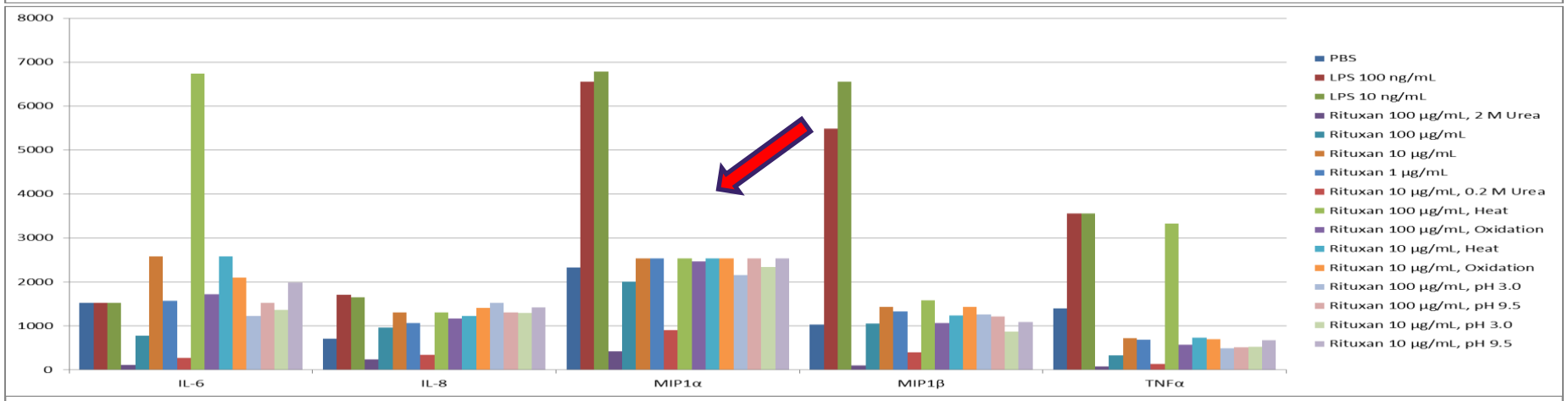


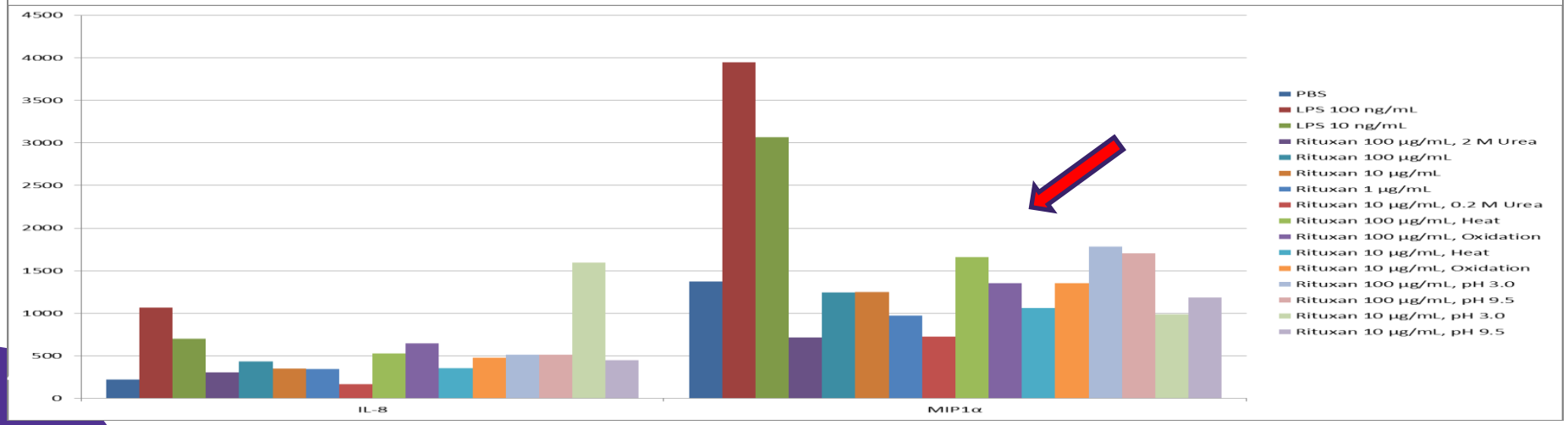
Only 11 of the 23 cytokines measured was shown here.

43

Donors 1, 2 and 3







5. Conclusions

- Antibody arrays were developed against 17 marketed Biologics and one for novel mAbs.
- Each antibody array provides a unique HOS signature for the mAb, reflecting its surface exposure and extent of exposure.
- The antibody array is sensitive, systematic and relatively high throughput.
- It correlates well with stability and bioassay data.
- It can detect changes that may not be detected with bioassays.
- It can be applied to many stages of biologics development, from cell line selection to product release and also used as an easy and accurate ID test.
- The xMAP format offers higher throughput and dynamic range, similar sensitivity and reduced cost as compared with the ELISA format and successfully applied in the bioprocess development.

Acknowledgement

Array Bridge: Michael Davies; Trevor Landon; Guofeng Fu.

MilliporeSigma: Wen-Rong Lie; Robert Keith; Xiao Qiang; Michael Godeny, Lawrence Rentoul.