AMGEN'S ATTRIBUTE BASED CONTROL STRATEGIES

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Attribute based control strategies

Attribute based control strategies are a fundamental element of our QBD approach, and are founded on controlling product quality attributes to targeted levels determined by a risk and science-based understanding of the impact of the attribute on safety and efficacy

The implementation of these strategies include:

- Optimal levels of in-process and specification testing
- Detailed understanding of product analytics and the application of new technologies to drive attribute-based control
- Leveraging of prior and platform knowledge in the determination of attribute impacts and target levels

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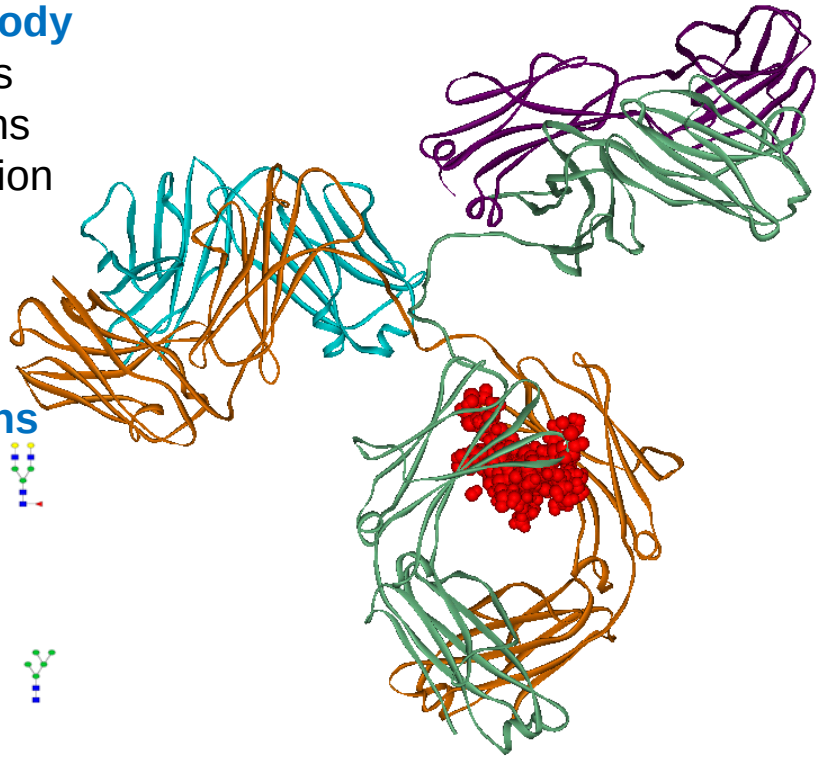
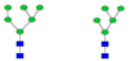
Biologics have numerous product quality attributes
Attribute based control is designed for identification, and appropriate control of each of the Critical Quality Attributes of a molecule

Monoclonal Antibody

~ 1300 amino acids
4 polypeptide chains
N-linked glycosylation
~ 150 kDa



Glycan modifications

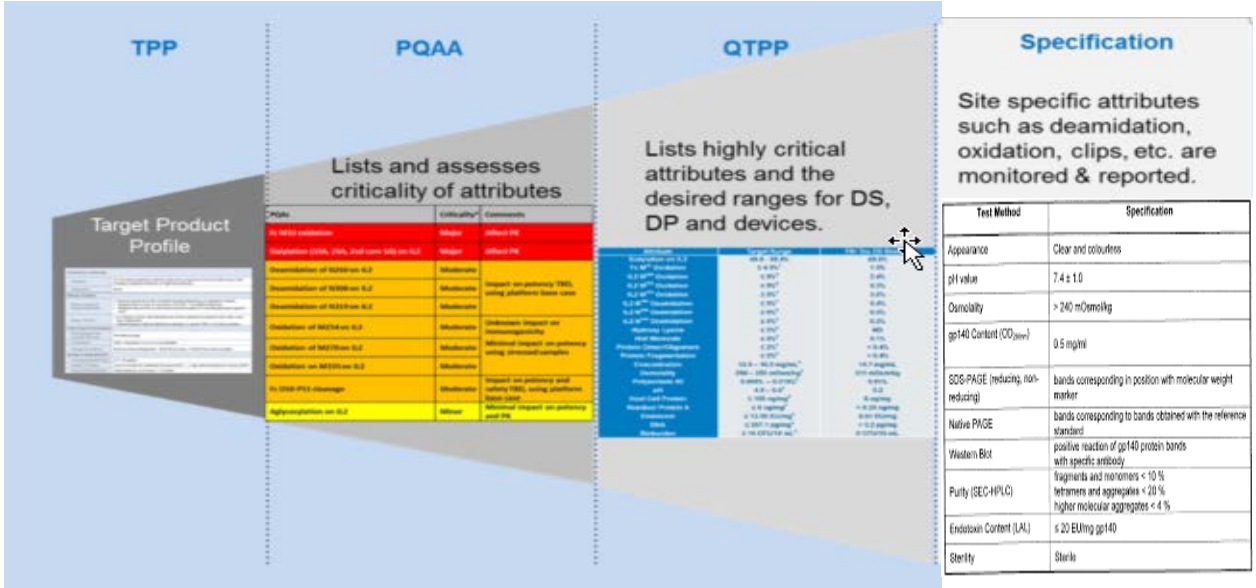
- G0, G1, G2 
- Core fucosylation
- Sialylation 
- High mannose 
- etc.

Peptide modifications

- Deamidation
- Succinimide
- Oxidation
- Glycation
- C-terminal variants
  - HC- Lys
  - HC-ProAmide
- N-terminal variants
  - Pyro Glu
- Amino acid substitution
- Truncation
- Half molecules
- Disulfide isoforms

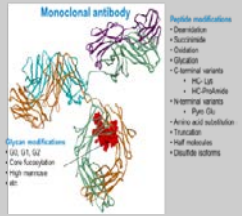
Attribute based control

- Attribute based control begins with the clinical and commercial goals of the Target Product Profile (TPP)
- A Product Quality Attribute Assessment (PQAA) is used to assess the impact of the product quality attributes on safety and efficacy, and determine the Critical Quality Attributes (CQAs)
- A Quality Target Product Profile (QTPP) is used to establish the target levels of the CQAs that will meet the goals of the TPP
- This information is used during product candidate selection and throughout the product lifecycle
- The QTPP is ultimately aligned with, though not identical to the final product Specifications



Product Quality Attribute Assessment (PQAA): identify attributes & impact
Quality Target Product Profile (QTPP): setting appropriate target ranges

Target Product Profile → ***Identifying Attributes*** → ***Scoring impact on safety and efficacy*** → ***Target ranges***



Product Quality Attribute Assessment

Quality Target Product Profile

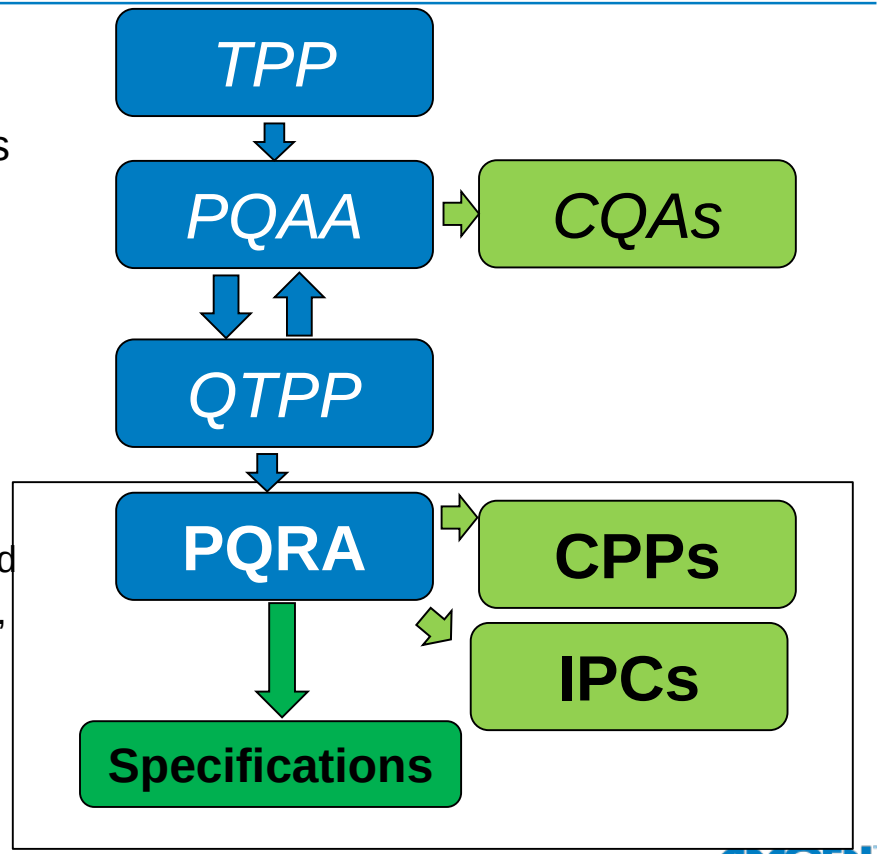
CDR modifications	Oxidation, Deamidation, Isomerizatio (molecule specific)	• Loss of potency	• Low, < x %
Fc binding regions	Methionine oxidation	• PK and efficacy	• Low, < x % ± y%
Glycan structure	High mannose variants (IgG class)	• PK and efficacy	• Low, < x % ± y%
	Sialylation	• PK	• high x- y%
Other backbone modifications and aggregated forms	Disulfide variants (IgG2, IgG4)	• Potency	• Depends on criticality
	Truncated/clipped forms	• Potency and PK due to missing functional regions	• high, < x%
	Host Cell Protein	• Immunogenicity	• xppm

Product Quality Risk Assessment

A Product Quality Risk Assessment (PQRA) is also applied for each PQA across each unit operation, to de-risk where the process impacts the attributes

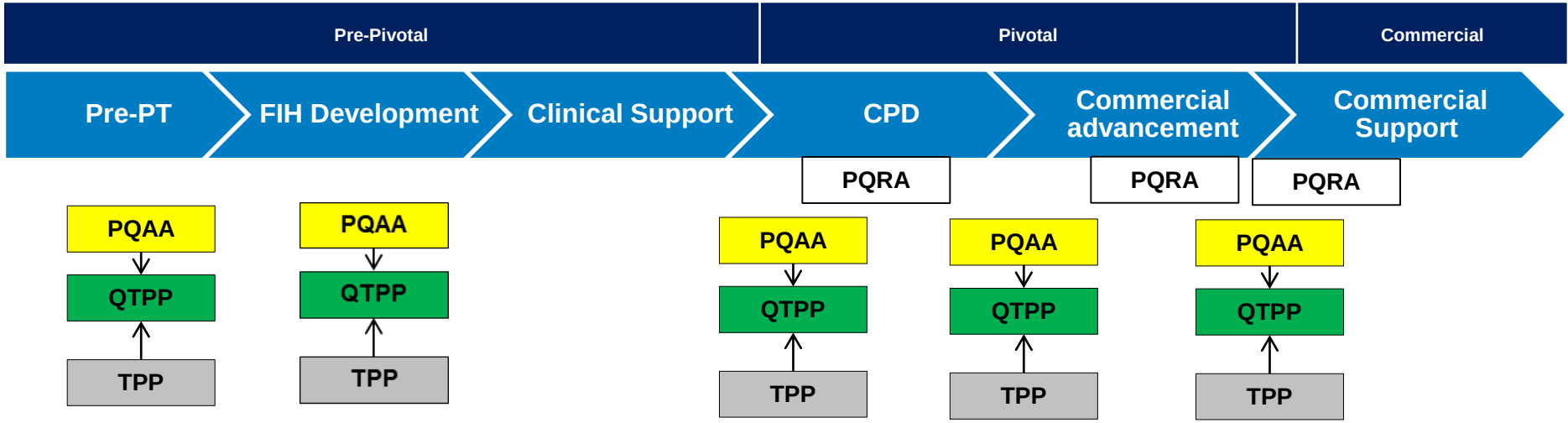
The PQRA is used iteratively with prior and accumulated process knowledge to help establish the integrated control strategy. The outcome of this combined process understanding results in:

- Critical process parameters determined at each unit operation where a PQA is impacted
- In-process, specification and stability testing, which reduce or mitigate risks identified by the PQRA to attribute control



End-to-end lifecycle: TPP,PQAA, QTPP and PQRA

As the clinical and commercial product targets evolve during the product lifecycle, and knowledge is accumulated on the product and process, the TPP, QTPP and PQAA are adjusted and aligned against each other, and eventually along with the PQRA



TPP – Target Product Profile
QTPP – Quality Target Product Profile
PQAA – Product Quality Attribute Assessment
PQRA – Product Quality Risk Assessment

These assessments drive process development focus and improvement

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Target Product Profile

- Attribute base control begins with the clinical and commercial goals of the Target Product Profile (TPP)

Attribute		Performance
Order of Entry/MoA		1st in Class
Efficacy	Symptom day reduction	• Reduction in XX at AA wks vs placebo = -YY days
	Responder rate	• BB% of patients experience CC% reduction in Z at 12wks
	Sub-populations	• Baseline characteristics in label state significant proportion of patients included in pivotal studies had previously had an inadequate response, loss of response, or intolerance to prior treatments
	Reduction in meds	• Reduction in use & type of medications
Tolerability		• Tolerability profile comparable to in a single head to head trial
Safety/Side Effects		• Safety profile without any major safety findings • Evidence of long term safety at time of launch
Dosing & Admin	Dosing & Admin	Single QM dose, SC injection
	Device	Autoinjector

Product Quality Attribute Assessment

The Product Quality Attribute Assessment (PQAA) is applied to the product quality attributes of the product to determine the Critical Quality Attributes (CQAs)

- Each PQA is scored independently for Safety and for Efficacy, resulting in a Overall Severity Score for the attribute
- The determination of scores relies heavily on prior and platform knowledge
- Safety is scored in terms of both immunogenic and non-immunogenic risks, while Efficacy scores consider PK and Potency impacts.
- Safety concerns will determine a CQA regardless of efficacy scoring
- Scores range from 1 (low impact) to 9 (high impact), with overall scores ≥ 5 generally considered CQAs

	Product Quality Attribute	Immunogenicity - Reduce Risk	Non-immune Safety - Reduce Risk	PK - Long PK desired	Potency-Loss Target High, Consistent Potency (No Limit)	Potency-Gain Target High, Consistent Potency (No Limit	Directional Goal
Purity-Product related	Met Ox (CDR)	5	NA	NA	5	NA	Low
	Met Ox (non CDR)	5	NA	7	1	NA	Controlled
	Mutation Misincorp	5	NA	5	1	NA	Controlled
Glycosylation	Fucosylation	1	7	1	NA	9	High
	Galactosylation	1	7	1	1	1	Controlled
	High Mannose	1	7	7	NA	7	Low

The scientific understanding of the CQA impact on safety and efficacy based on prior knowledge may be sufficiently comprehensive to allow limits to be set independently of clinical experience

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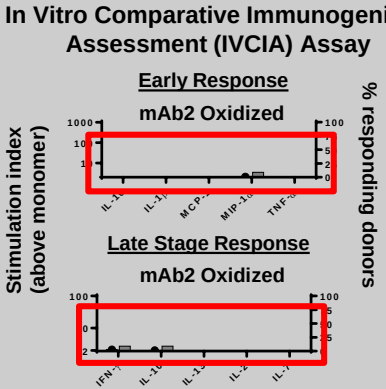
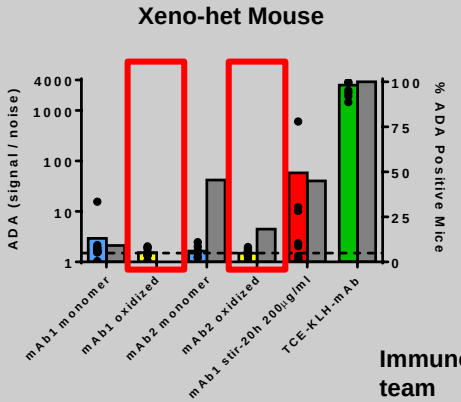
Appropriate Specifications are Critical to Integrated Control Strategies

Experimental data indicate that Met Ox no apparent impact on Safety or Efficacy





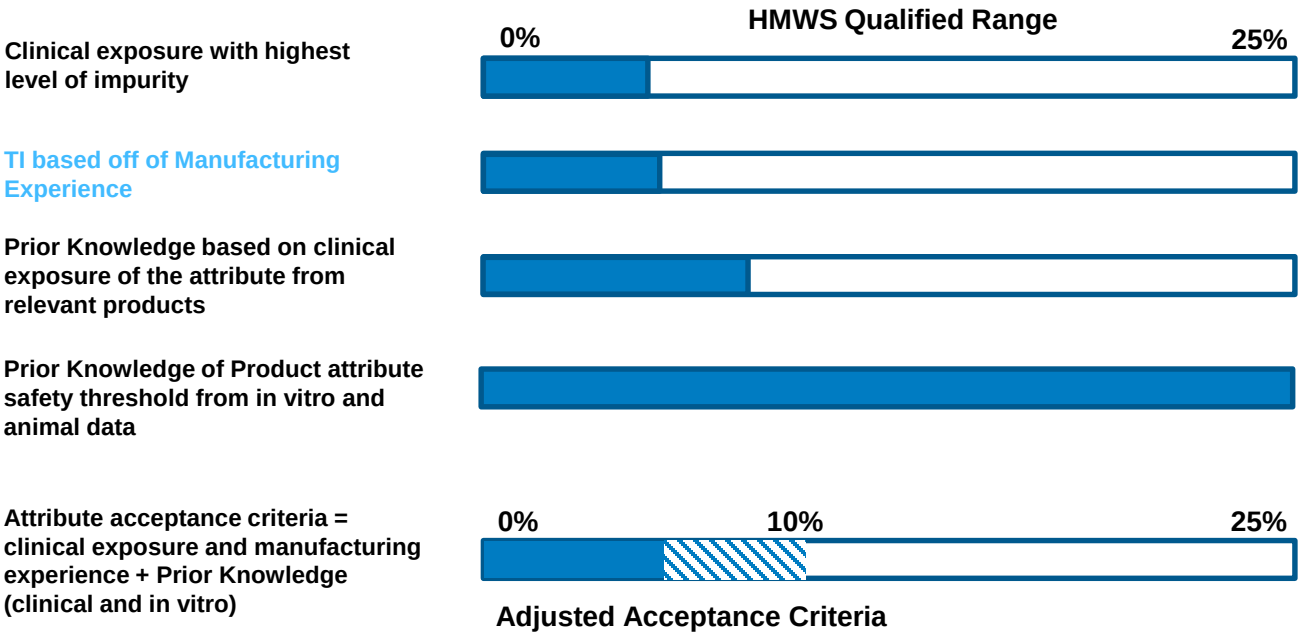
Immunogenicity CQA team

Conclusions:

- Safety: Met oxidation does not appear to increase immunogenicity risk as shown by the in vitro cell-based assays and the in vivo Xeno-het mouse model
- Clearance: Oxidation at the conserved Fc met 252 and 428 under reasonable conditions has negligible impact on FcRn binding and subsequent PK clearance (Stracke et al., mAbs, 2015 6:5, 1229-1242)

RISK AND SCIENCE BASED SPECIFICATIONS FOR MET OX AND SIMILAR ATTRIBUTES SHOULD NOT BE LIMITED TO CLINICAL EXPERIENCE WHERE PRIOR KNOWLEDGE INDICATES LOW RISK

ATTRIBUTE BASED SPECIFICATIONS SHOULD BE ESTABLISHED BY APPLICATION OF BOTH CLINICAL EXPERIENCE AND APPLICABLE PRIOR KNOWLEDGE



THIS APPROACH WOULD ACHIEVE KNOWLEDGE AND EXPERIENCE BASED SPECIFICATIONS

Quality Target Product Profile

- The QTPP is used to establish the target levels of CQAs determined by the PQAA, that will meet the goals of the TPP in terms of safety, efficacy, PK, dosing, etc.
- The PQAA and QTPP are applied iteratively through the lifecycle of a product, as more knowledge is accumulated on the product and the process

Category	Attribute	Target Range	Current Observed Range
Strength	Concentration	126 – 154 mg/mL	131 – 149 mg/mL
Quality	HC Asp Isomerization	≤ 2%	0.1 – 0.5%
	LC Trp Oxidation	≤ 5%	0.1%
	HC Met Oxidation	≤ 5%	0.3 – 0.9%
	HC Met Oxidation	≤ 5%	0.4%
	Met Oxidation	1% – 7%	2.5 – 4.1%
	Met Oxidation	≤ 5%	0.7 – 1.6%
	High Mannose Glycans	2% – 12%	6.2 – 8.5%
	Protein Dimer/Oligomers (SEC HMW)	≤ 1%	0.4 – 0.6%
	Protein Fragmentation (rCE LMW+MMW)	≤ 1%	< 0.6%
	Glycation (LC K)	≤ 5%	0.8 – 1.5%
	Hydroxylysine (HC K)	≤ 2%	< 0.1%
	Hydroxylysine (HC K)	≤ 2%	1.0 – 2.0%
	Osmolality	250 – 350 mOsm/kg	301 – 312 mOsm/kg
	Polysorbate 80	0.005% – 0.015%	0.009 – 0.013%
	pH	4.9 – 5.5	5.1 – 5.2
Safety	Host Cell Protein	≤ 100 ppm	20 – 49 ppm
	Residual Protein A	< 6 ppm	< 1 ppm
	Endotoxin	≤ 0.25 EU/mg	≤ 0.0022 EU/mg
	Bioburden	≤ 10 CFU/10 mL	0

Product Quality Risk Assessment (PQRA)

A Product Quality Risk Assessment is applied - combined with process understanding - for each CQA/PQA across each unit operation, to assess where the process impacts the attributes

Quality Attribute \ Unit Operation	cDNA	Clone/cell line	Cell banking	Expansion in Flasks	Vial Thawing	Bioreactors	Production Bioreactor	Harvest	Harvest Pool Hold	Column 1	Column 1 Pool Hold	Low pH / VI	Filtered VI Pool Hold	Column 2	Column 2 Pool Hold	Column 3	Column 3 Pool Hold	Viral Filtration	VF Pool Hold	UF/DF	UF/DF Pool Hold	Drug Substance
Oligomer		Low					Medium		Low	Low	Low	Low	Low	Low	Low	Low	Low	Low	Low	Low	Low	
Aggregates		Low					Medium		Low	Medium	Low	High	Low	Low	Low	Low	Low	Low	Low	Medium	Low	
CDR Trp-ox							Low		Low		Low		Low		Low		Low		Low		Low	
C-term lys							Low									Low	Low					
Deamidation - CDR							Low		Low						Low							
Deamidation - non-CDR							Low		Low					Low	Low							
Fragmentation							Low	Medium	Low	Low												
Free –SH, Cys Adducts	Low						Low	Medium	Low													
Glycation							Low		Low						Low		Low					
HC C-term proline amidation		Low					Low															
Hydroxylysine		Low					Low															
IgG2 Disulfide Isoforms							Low															
Met Ox CDR							Low	Low														
Met Ox non-CDR							Medium	Medium														Low
N-term Pyro Glu							Low															
N-term signal seq variants	Low	Low					Low															
Thioether							Low															
Trisulfide							Low															
Fucosylation							High															
Galactosylation							High															
High Mannose Glycan Species		Low					High															
Non-consensus Glycosylation							High															
Non-glycosylated HC							High															
Sialic Acid							Low															
Unusual Fc glycans							Low															
CHOP							Low	Low		High		Low		Low		Low						
Residual host cell DNA							Low	Low		Low				Medium		Low						
Residual Protein A										Low				Low		Low						
Appearance																				Low	Low	
Bioburden			Low	Low		Low	Low	High	Medium	Medium	Medium	Medium	Medium	Medium	Medium	Medium	Medium	Medium	Medium	Medium	Medium	Low
Clarity																				Low	Low	
Color	Low																			Low	Low	
Endotoxin				Low		Low	Low	Medium	Low	Low	Low		Low	Low	Low	Low	Low		Low	Low	Low	
Osmolality																				Low		
pH																				Low		
Product Conc.																				Low		

Product Quality Risk Assessment

The PQRA evaluates each PQA using an FMEA approach, including:

- **Qualitative impact of the unit operation (UOP) on the attribute**
- **Control elements associated with the UOP including procedural controls, raw material controls, and testing controls including characterization and/or comparability**
- **Occurrence score for the UOP and associated risk level**
- **Detection at the UOP, where detection score accounts for capability and stringency of detection, and overall risk accounts for detection downstream of each UOP**

Quality Attribute:	Potential Adverse Impact:	Severity Score:
Oligomer - HMWS ≤ tetramer	Minor impact to potency	7

	Unit Operation	Correlation (↑, ↓ or testing only)	Control Elements															Occurrence			Preliminary Hazard Risk Level	Detection at Unit Operation				Downstream Detection		Overall Unit Operation Risk Level
			Procedural Controls	Raw Material and Components	Characterization Only	Clinical In-process (PD, Clin Mfg)	Process/Product Monitoring	IPC (Action Limit)	IPC (Reject Limit)	Clinical INV Limits	DSI Release Spec	DS Release Spec	DP Release Spec	DS Stability	DP Stability	Validation (DS, DP, Transport, etc)	Comparability	Occurrence Supporting Information	Occurrence Decision Tree Code	Occurrence Score		Detection Method	Capability (n)	Stringency (i)	Detection Score	Detected downstream (if yes, list step)?	Overall Detection Score	
12	Filtered VI Pool Hold	↑	x		x		x									x		HMP pool hold studies	e	1	Low	SEC (MET-xxxxx)	9	5	7	Yes, Column 2 Pool Hold	1	Low
13	Column 2	↓	x		x													Development studies and confirmation runs	e	1	Low	SEC (MET-xxxxx)	9	9	9	Yes, Column 2 Pool Hold	1	Low
14	Column 2 Pool Hold	↑	x		x		x									x		HMP pool hold studies	e	1	Low	SEC (MET-xxxxx)	9	5	7	Yes, Column 3 Pool Hold	1	Low
15	Column 3	↓	x		x													Development studies and confirmation runs	J	5	Medium	SEC (MET-xxxxx)	5	9	7	Yes, Column 3 Pool Hold	1	Low
16	Column 3 Pool Hold	↑	x		x			x								x		HMP pool hold studies	J	5	Medium	SEC (MET-xxxxx)	5	3	4	Yes, UFDF Pool Hold	1	Low
17	Viral Filtration	↑	x		x											x		Development studies and confirmation runs	J	5	Medium	SEC (MET-xxxxx)	9	5	7	Yes, UFDF Pool Hold	1	Low
18	VF Pool Hold	↑	x		x		x											HMP pool hold studies	f	3	Medium	SEC (MET-xxxxx)	9	9	9	Yes, UFDF Pool Hold	1	Low
19	UF/DF	↑	x		x													Development studies and confirmation runs	f	3	Medium	SEC (MET-xxxxx)	5	9	7	Yes, UFDF Pool Hold	1	Low
20	UF/DF Pool Hold	↑	x		x		x									x		HMP pool hold studies	f	3	Medium	SEC (MET-xxxxx)	5	5	5	Yes, DS Release and Stability	1	Low
	Quality Attribute Overall Risk Level																											Low

Product Quality Risk Assessment

- **The level of risk of to the control of a CQA can be reduced by adding specific in-process, stability or specification testing or improving process control**
- **Testing may include no tests, where the PQA is controlled at low risk without testing, in-process testing only where specification testing is not required, to specification and stability testing where appropriate**
- **The introduction of new testing technologies such as MAM* allow for PQA specific testing, rather than category base testing (e.g., acidic species)**

***MAM is a peptide map/mass spectrometric methodology employed by Amgen to assay multiple specific PQAs**

Quality Attribute:	**Potential Adverse Impact:**	**Severity Score:**
Oligomer - HMWS ≤ tetramer	Minor impact to potency	7

	Unit Operation	Correlation (↑, ↓, or testing only)	Control Elements: Procedural Controls	Raw Material and Components	Characterization Only	Clinical In-process (PD, Cln Mfg)	Process Control Monitoring	IPC (Action Limit)	IPC (Reject Limit)	Clinical INV Limits	DSI Release Spec	DS Release Spec	DP Release Spec	DS Stability	DP Stability	Validation (DS, DP, Transport, etc)	Comparability	Occurrence: Occurrence Supporting Information	Occurrence Decision Tree Code	Occurrence Score	Preliminary Hazard Risk Level	Detection at Unit Operation: Detection Method	Capability (n)	Stringency (i)	Detection Score	Downstream Detection: Detected downstream (if yes, list step)?	Overall Detection Score	Overall Unit Operation Risk Level
12	Filtered VI Pool Hold	↑	x		x		x									x		HMP pool hold studies	e	1	Low	SEC (MET-xxxxx)	9	5	7	Yes, Column 2 Pool Hold	1	Low
13	Column 2	↓	x		x													Development studies and confirmation runs	e	1	Low	SEC (MET-xxxxx)	9	9	9	Yes, Column 2 Pool Hold	1	Low
14	Column 2 Pool Hold	↑	x		x		x									x		HMP pool hold studies	e	1	Low	SEC (MET-xxxxx)	9	5	7	Yes, Column 3 Pool Hold	1	Low
15	Column 3	↓	x		x													Development studies and confirmation runs	J	5	Medium	SEC (MET-xxxxx)	5	9	7	Yes, Column 3 Pool Hold	1	Low
16	Column 3 Pool Hold	↑	x		x			x								x		HMP pool hold studies	J	5	Medium	SEC (MET-xxxxx)	5	3	4	Yes, UFDF Pool Hold	1	Low
17	Viral Filtration	↑	x		x											x		Development studies and confirmation runs	J	5	Medium	SEC (MET-xxxxx)	9	5	7	Yes, UFDF Pool Hold	1	Low
18	VF Pool Hold	↑	x		x		x											HMP pool hold studies	f	3	Medium	SEC (MET-xxxxx)	9	9	9	Yes, UFDF Pool Hold	1	Low
19	UF/DF	↑	x		x													Development studies and confirmation runs	f	3	Medium	SEC (MET-xxxxx)	5	9	7	Yes, UFDF Pool Hold	1	Low
20	UF/DF Pool Hold	↑	x		x		x									x		HMP pool hold studies	f	3	Medium	SEC (MET-xxxxx)	5	5	5	Yes, DS Release and Stability	1	Low
	Quality Attribute Overall Risk Level																											Low

Testing is reduced to the level necessary, in alignment with the risks, with the elimination of redundant or non-value added testing

Critical Process Parameters

Critical process parameters are determined at each unit operation where the CQA is impacted, based on process knowledge

- **ICH Q8: Critical Process Parameter: A process parameter whose variability has an impact on a critical quality attribute and therefore should be monitored or controlled to ensure the process produces the desired quality.**
- **The determination of process parameter impacts on CQAs can be derived from prior or platform knowledge, DOE studies and/or in-silico modelling.**
- **The impact ratio method is computed as the change in a CQA from the midpoint to the limit of the process parameter acceptable range (AR) divided by the difference between the CQA value at the midpoint of the PP acceptable range and the acceptable limit of the CQA at the process step.**

$$\text{Impact Ratio} = \frac{\text{CQA Value at Process Parameter limit} - \text{CQA Value at the Process Parameter Midpoint}}{\text{CQA Limit} - \text{CQA Value at the Process Parameter Midpoint}}$$

- **A process parameter with an impact ratio >0.2 is considered a CPP, based on the determination that a 20% shift in the CQA across the PP acceptable range is significant and identifies a CPP**

CPPs are not based on failure of an attribute across the process parameter range

Example: mAb non-CDR Fragmentation PQAA/QTPP

The PQAA for this specific fragmentation species might indicate an Overall Severity Score of 5 making this species a CQA

Quality Attribute	Attribute Information	Safety		Efficacy		PQA Severity Ranking
		Immuno-genicity Risk	Non-Immune Safety Risk	PK	Potency	
Non-CDR Fragmentation	1 potential DP site in Fc	5	N/A	5	5	5

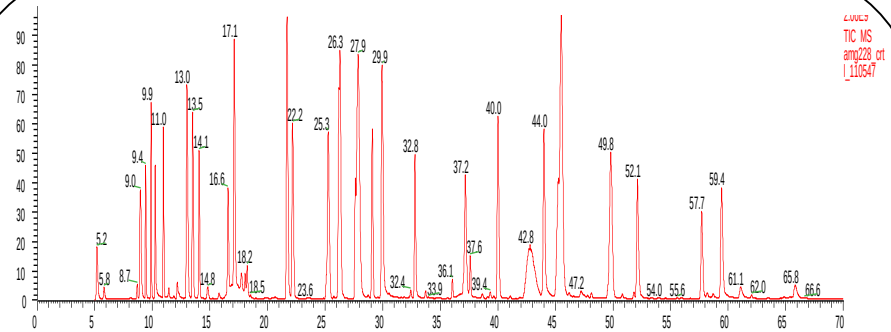
The QTPP range for non-CDR fragmentation for a typical product might be ~1% in order to ensure that at that the target level of dosing there is no impact on S&E

Attribute	Target Range
Non-CDR Fragmentation	**0 - 1%**

However, the typical clipped species assay (rCE-SDS) cannot distinguish between the different variants of clipped species. With a typical approach we cannot decouple control of this CQA from control of other attributes

Attribute specific control is enabled with methodologies such as the Multi Attribute Method

- MAM can directly identify and quantify specific PQAs, which enables Amgen to design a more attribute specific QTPP
- With regards to clipped species, MAM can identify the level of specific clips which are CQAs, distinguishing from those clipped species which are not, allowing detailed CQA control



Attribute	Residue	Peptide	Structural Region	%
Deamidation	N53	LC 3	LC CDR1	6.2
Oxidation	M39	H3	HC CDR1	0.2
	M113	H12	HC CDR3	3.8
	M253	H23	Fc	1.9
	M431	H44	Fc	0.9
Mannose glycan	N301	H21	Fc	10.2
Clips	D276/P277	H24	Fc	0.3

- MAM enables detection and quantification of specific product quality attributes

Example: non-CDR Fragmentation PQRA

- The PQRA for Drug Substance (DP not shown) indicates potential formation and/or removal at a number of steps
- Multiple controls are in place across the process
- In-process testing using MAM is included at the VI pool, which reduces the control risk
- There is no other testing - this is considered the optimal level of testing for the CQA

Quality Attribute: Clipped Species (D276/P277)

Potential Adverse Impact: Potential impact to immunogenicity or efficacy

Severity Score: 5

Unit Operation	Correlation (↑, ↓ or testing only)	Control Elements: Procedural Controls	Raw Materials and Components	Characterization Only	Clinical In-process Mon (Can Mfg)	Process Control Monitoring	IPC (Reject Limit)	Clinical INV Limits	Clinical Monitoring Program (DS, DP)	DSI Release Spec	DS Release Spec	DP Release Spec	DS Stability	DP Stability	Validation (DS, DP, Transport, etc)	Comparability	Occurrence: Occurrence Supporting Information	Occurrence Decision Tree Code	Occurrence Score	Preliminary Hazard Risk Level	Detection at Unit Operation: Detection Method	Capability (n)	Stringency (i)	Detection Score	Downstream Detection: Detected downstream (if yes, list step)?	Overall Detection Score	Overall Unit Operation Risk Level
Production Bioreactor	↑	x	x															K	5	Medium	No testing at this step	9	9	9	Yes, Column 1 Validation	4	Low
Harvest	↑	x	x															K	5	Medium	No testing at this step	9	9	9	Yes, Column 1 Validation	4	Low
Harvest Pool Hold	↑	x	x															K	5	Medium	No testing at this step	9	9	9	Yes, Column 1 Validation	4	Low
Column 1	N/A	x	x												x		Development studies and confirmation runs (2K and 300L)	J	5	Medium	No testing at this step	1	7	7	Yes, VI Pool	4	Low
Low pH / VI	↑	x	x														Development studies and confirmation runs (2K and 300L)	e	1	Low	MAM method (MET-XXXXX)	5	9	4	No	1	Low
Drug Substance	N/A		x												x	x	N/A				No testing at this step	1	7	7	No	4	
	Quality Attribute Overall Risk Level																										Low

Example: mAb non-CDR Fragmentation Critical Process Parameters (CPPs)

- Prior knowledge, process models, and DOE experiments provide a knowledge basis for assessing nCPPs and potential CPPs
- Based on impact ratio calculations, one CPPs impacting non-CDR fragmentation may be identified at the Viral Inactivation step

Unit Operation	CPP	Operating Range	Impact Ratio
VI Pool Hold	Time	60-90 min	0.4

Summary

- Amgen's approach to attribute based control relies on prior knowledge and product specific knowledge to identify CQAs and establish safe and efficacious target ranges
- Attribute based control requires a detailed product characterization and/or appropriate analytical technologies to enable attribute specific measurements
- A deep understanding of the product, and the impact of the process on CQAs, allows specific control of CQA levels to designed targets in order to achieve clinical and commercial goals

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ACKNOWLEDGEMENTS

- Jette Wypych
- Mike Abernathy
- Bob Kuhn
- Trent Munro
- Chantal Cazeault
- Andy Spasoff