BioAgilytix

Immunogenicity assays for new modalities

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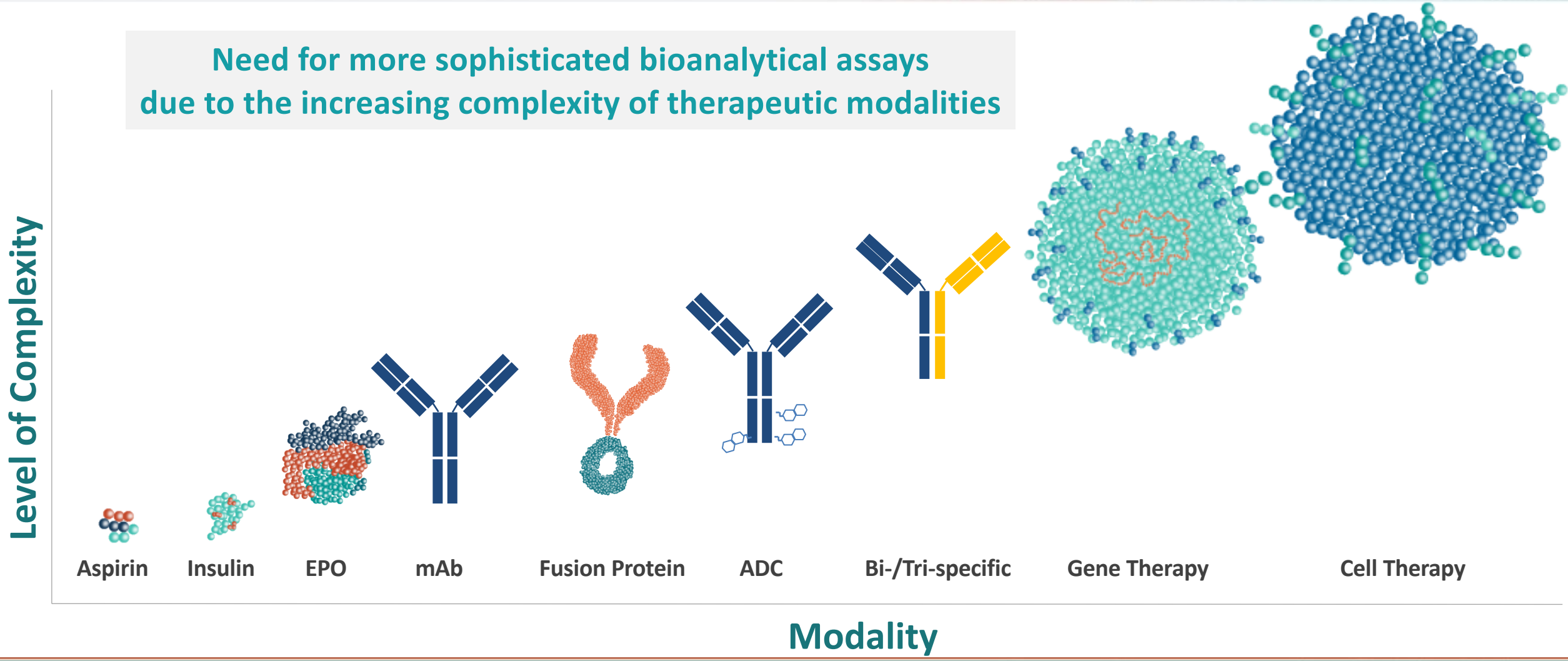
14th EBF Open Symposium

Evolution of Therapeutic Modalities

Need for more sophisticated bioanalytical assays due to the increasing complexity of therapeutic modalities

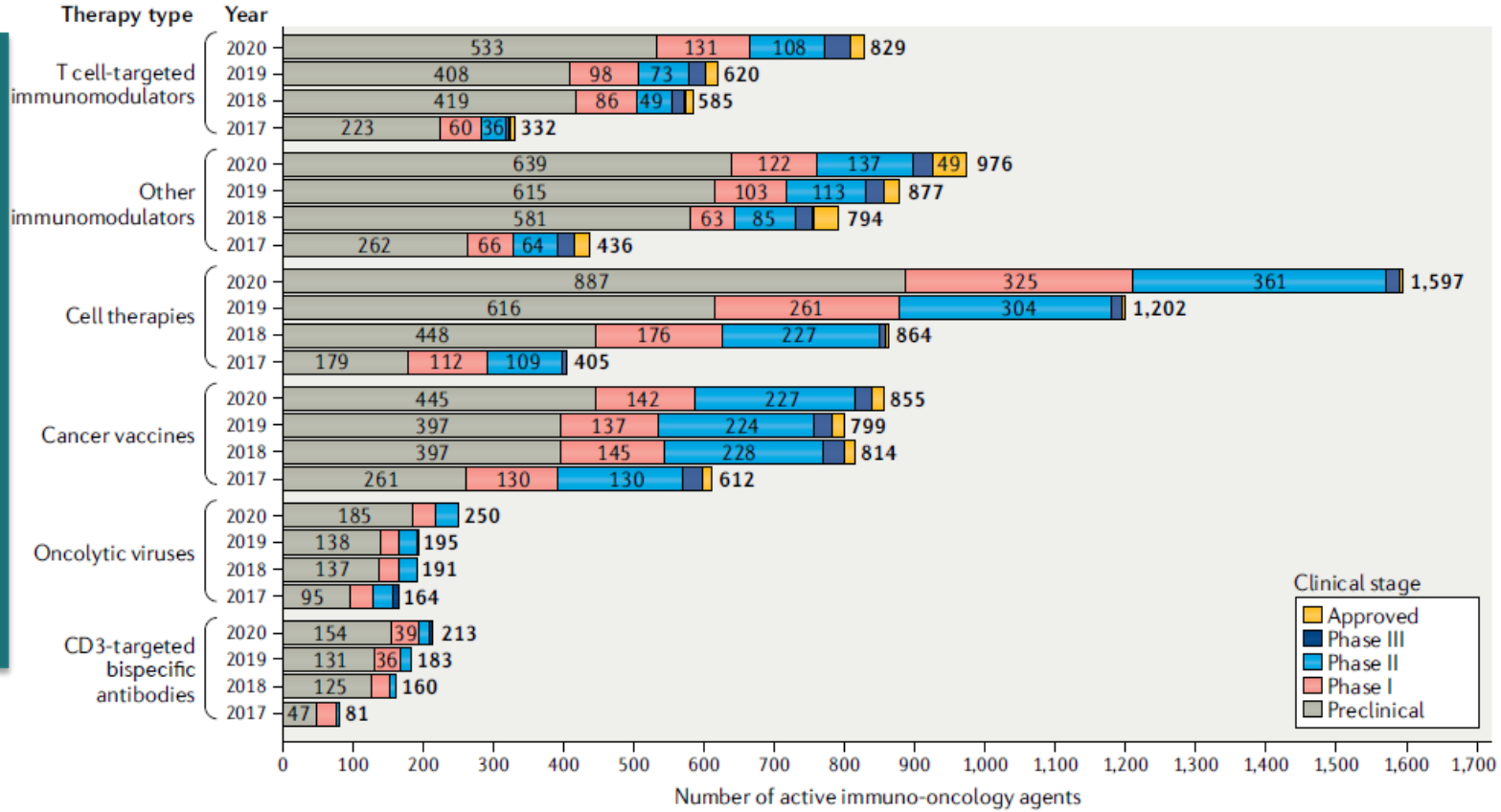


Immuno-Oncology Overview

4,720 immuno-oncology agents in the current global clinical pipeline

6,281 active clinical trials

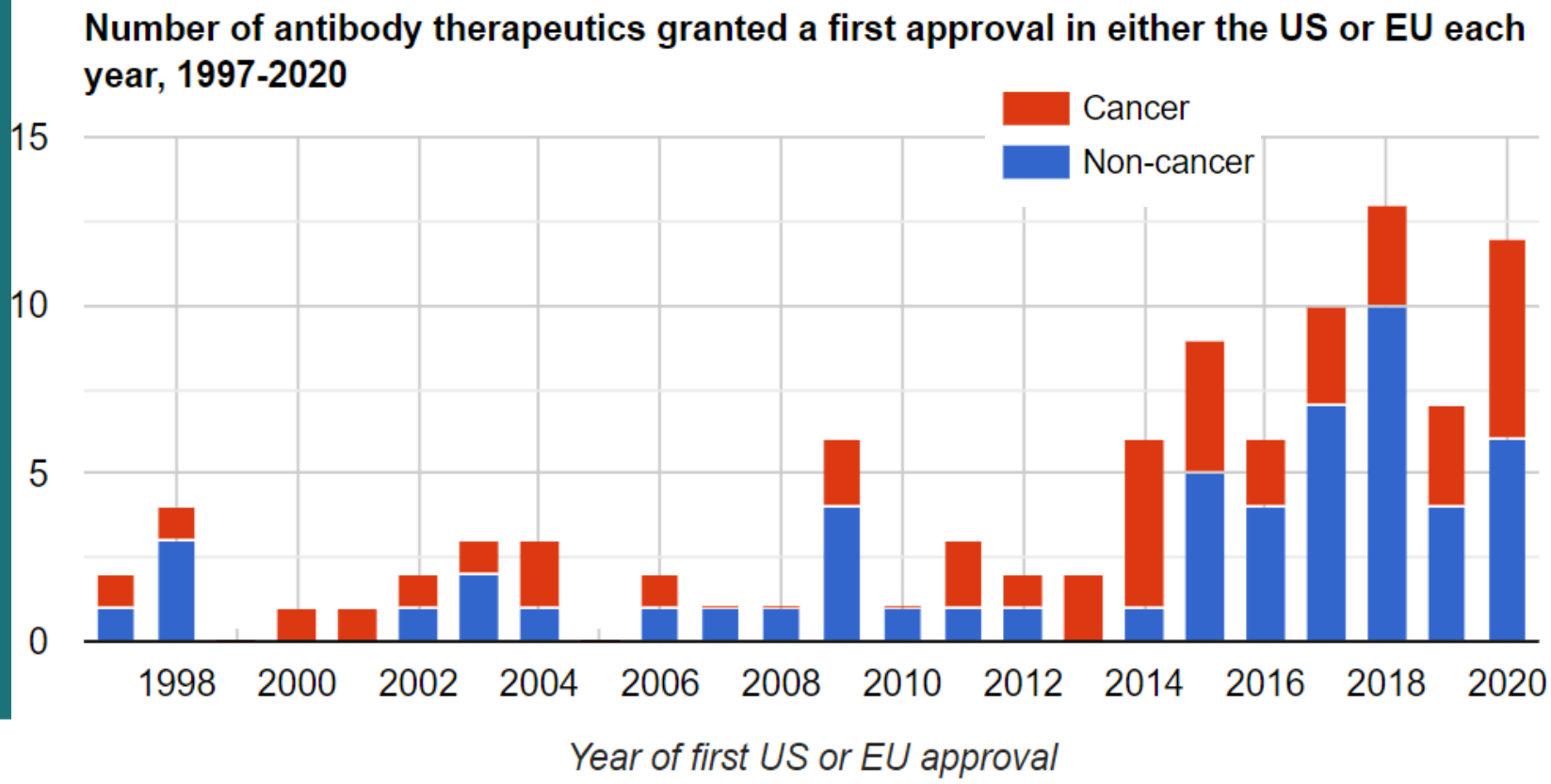
- CD3-Targeted Bispecific
- Cell Therapy
- T cell Targeted Immunomodulator



Antibody Therapeutics Overview

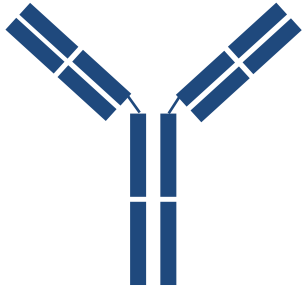
107 Therapeutic antibodies approved *)

- Currently 4 approved Bi-specific
  - Amivantamab (EGFR-cMET)
  - Catumaxomab (EpCAM – CD3)
  - Blinatumab (CD19 – CD3)
  - Emicizumab (FIX – FX)
  - Faricimab (VEGFA- Ang2) -review

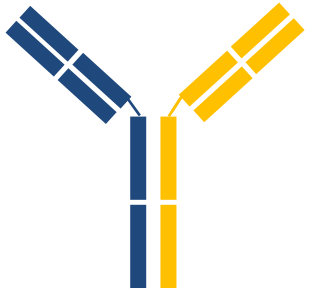


*) August 2021 – The Antibody Society

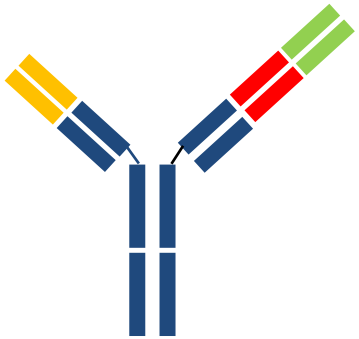
Drug Modality Structures



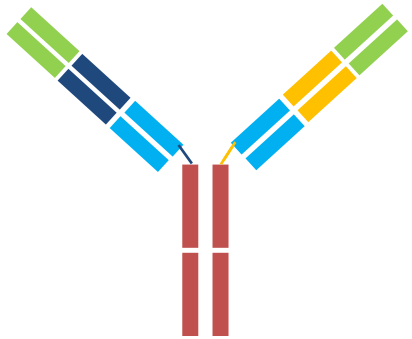
Monoclonal



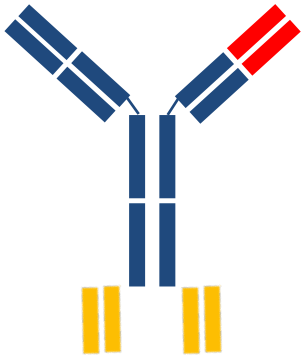
Bi-specific



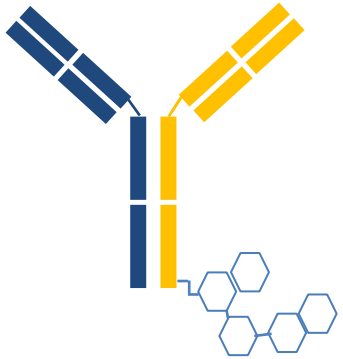
Tri-specific



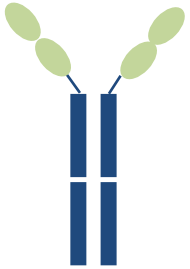
Bi-specific Dual variable domain (DVD)



Tri-specific



Antibody drug conjugate
ADC



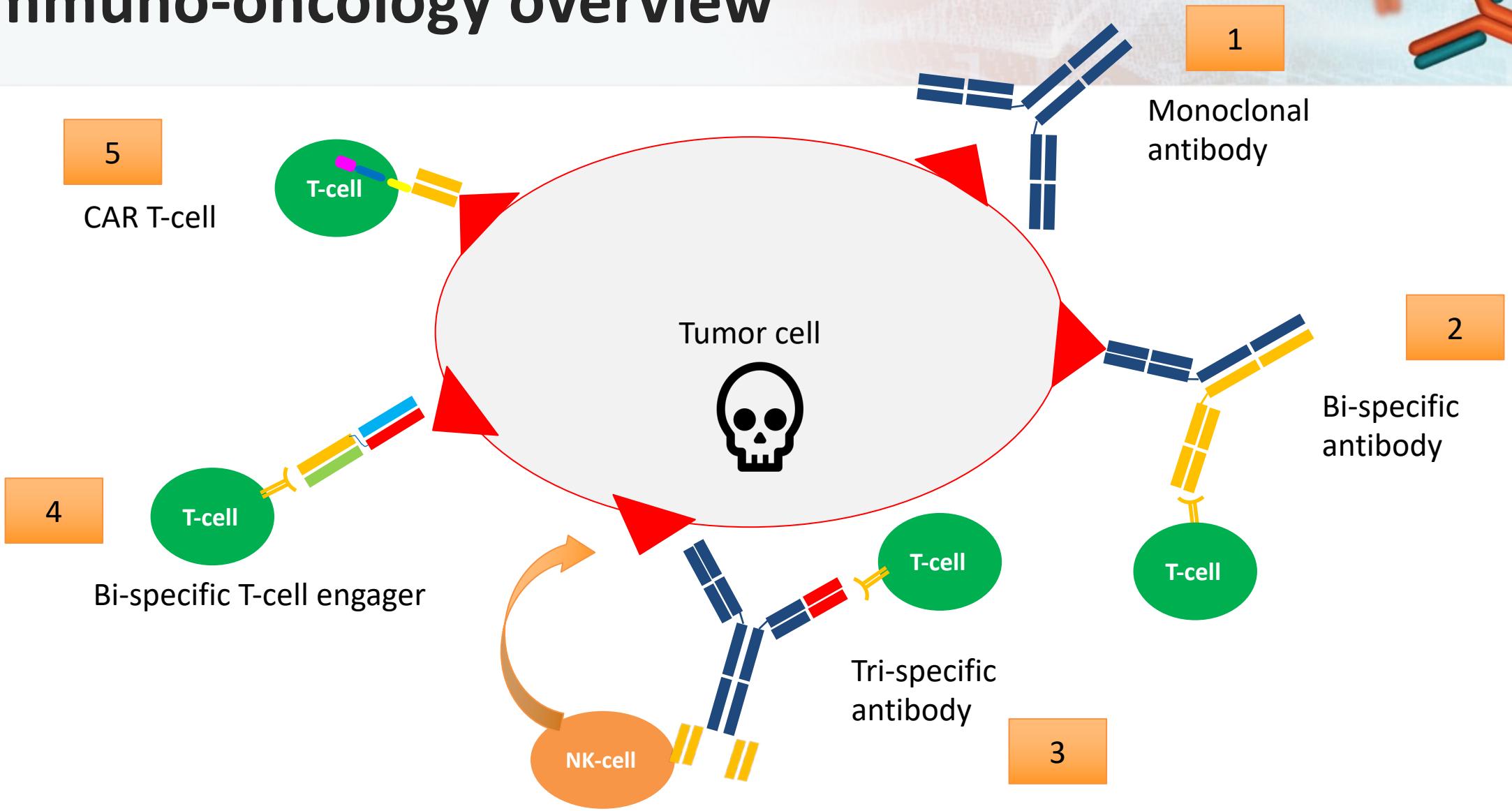
Peptibody

Fragment antigen binding
(Fab)2

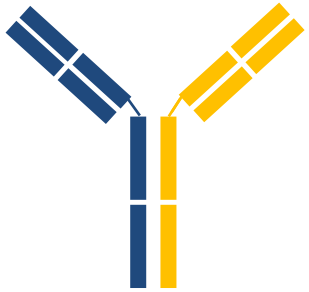
Single chain
Variable Fragment
ScFv

Bi-specific T-cell engager
BiTE

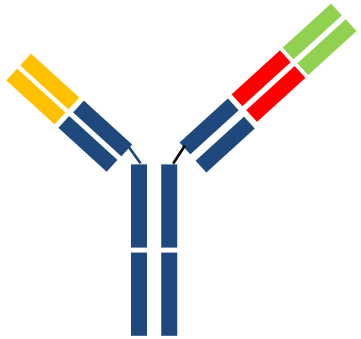
Immuno-oncology overview



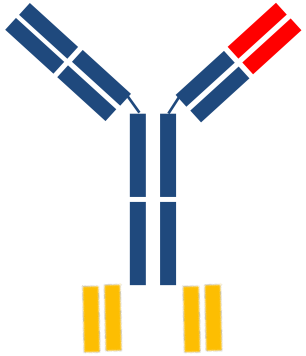
Immunogenicity of new modalities



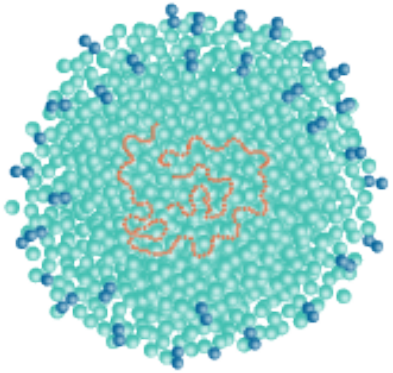
Bi-specific



Tri-specific



Tri-specific



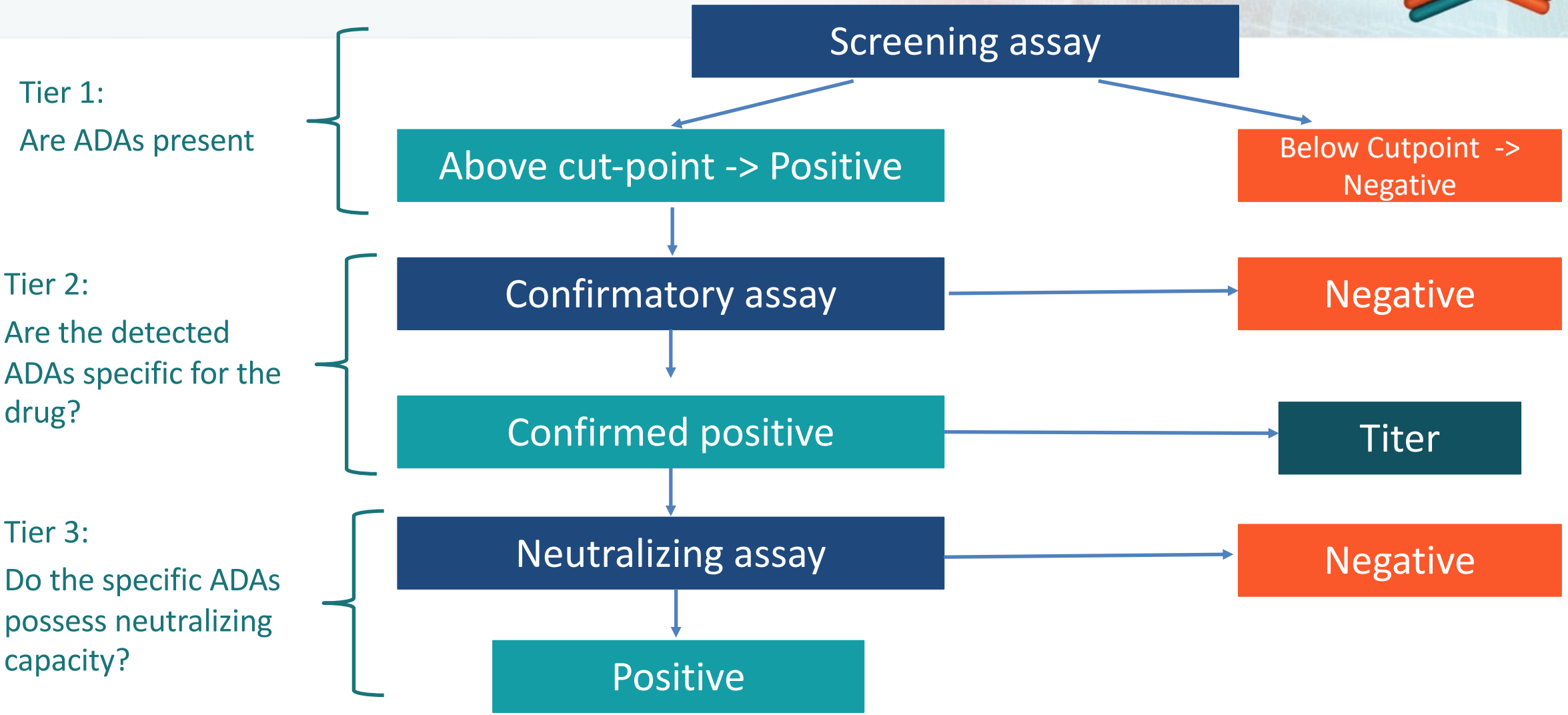
AAV vectors

- Sophisticated analytical techniques
- Needs for multiple assays
- High sensitivity
- Adherence to regulatory guidance

Validation parameters for ADA

- Specificity
- Assay cut-point
- Specificity (Confirmatory) cut-point
- Sensitivity
- Assay controls; precision; acceptance criteria
- Recovery
- Drug Interference
- Stability (short-term; freeze and thaw; long-term)
- Robustness

Assessment of ADA



ADA testing for Bi / Tri-specific Antibodies

- Test general validation parameters
- Follow the 3 Tiers approach
- Know the Target level & Drug Tolerance level

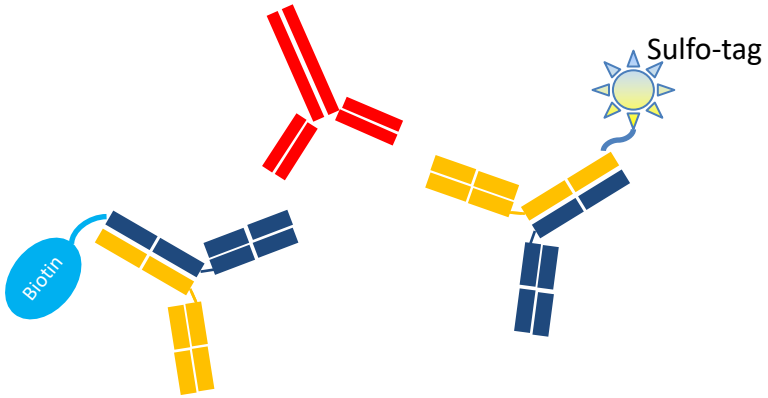
- Characterize the domain specificity based on the structure, the epitope, the linkers
- The need for domain-specific ADA and Nab assay can vary at different stages of drug development

Risk assessment early is essential in order to get started on the different assays that may be needed

ADA Testing Strategy for Bispecific

Screening Assay

Anti-Drug Antibody

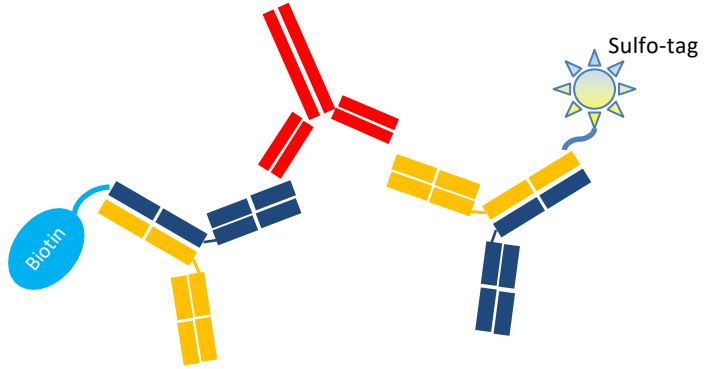


Consider a multi-tiered approach for specificity and characterization cut-point determination

Identifying the positive control can be a challenge

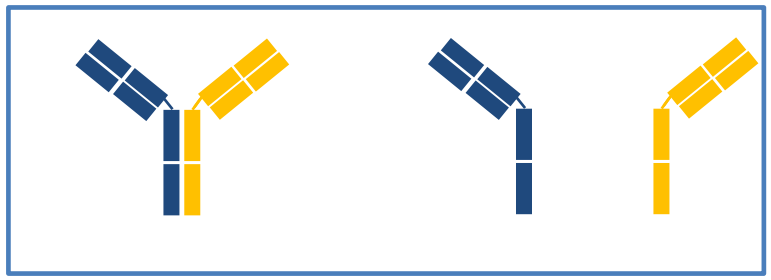
Confirmatory Assay

Anti-Drug Antibody



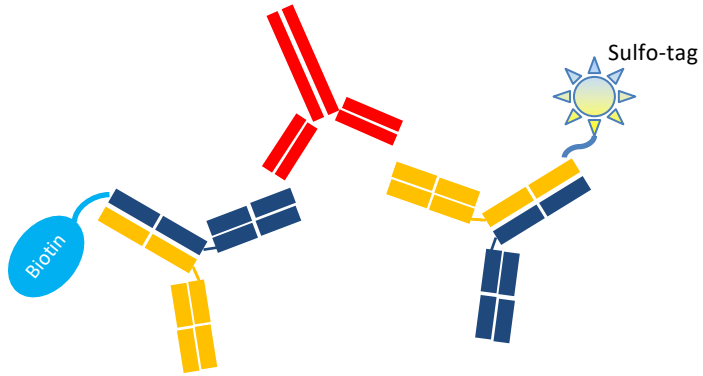
Cut-point per whole Ab and Domain

Confirm assay with Domain A and B



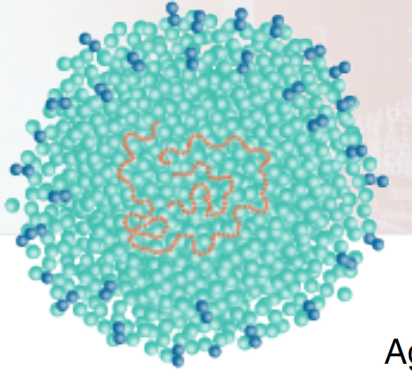
Titration Assay

Anti-Drug Antibody



Domain specific ADA characterization

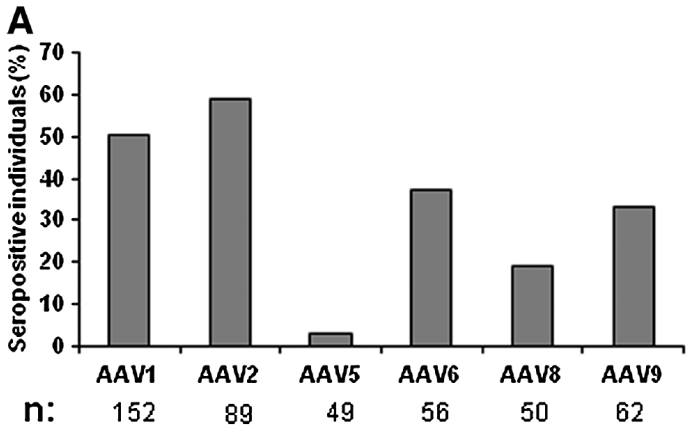
Gene Therapy



- Prevalence of anti-AAV antibodies
  - Pre-exisiting antibodies
- Considerations for Inclusion/Exclusion criteria
  - May limit transduction
  - Various approaches taken by sponsors
  - Currently, sponsors are beginning to rethink
- Redosing may not be possible
  - Prior exposure may limit the ability to redose with same or similar gene therapy candidate

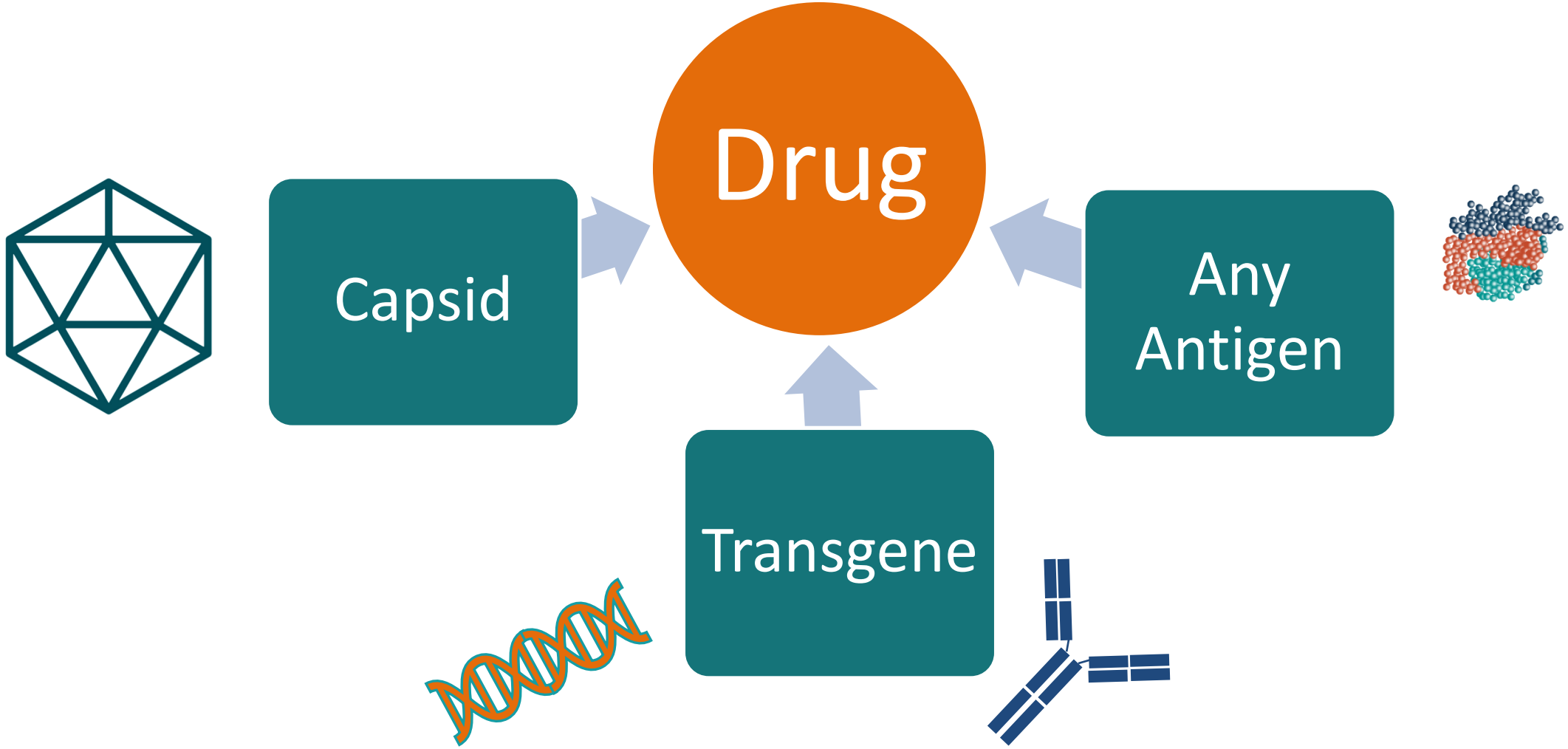
Prevalence of Serum IgG and Neutralizing Factors Against Adeno-Associated Virus (AAV) Types 1, 2, 5, 6, 8, and 9 in the Healthy Population: Implications for Gene Therapy Using AAV Vectors

Sylvie Boutin,[1] Virginie Monteilhet,[1] Philippe Veron,[1] Christian Leborgne,[1] Olivier Benveniste,[2] Marie Françoise Montus,[1] and Carole Masurier[1]

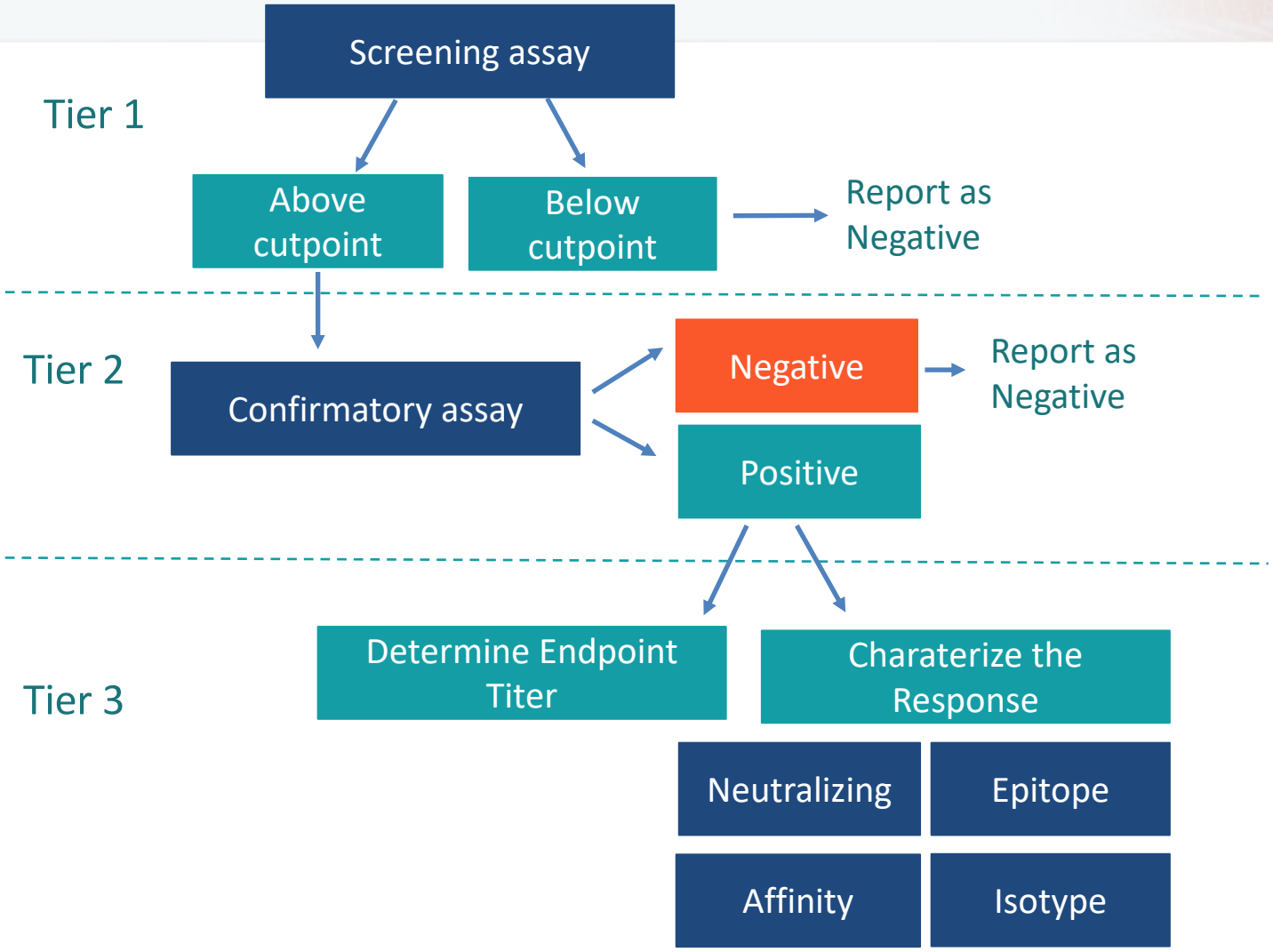


Human Gene Therapy vol 21, no. 6, 2010

What is the Drug in ADA testing?



Assessment of ADA –Gene Therapy



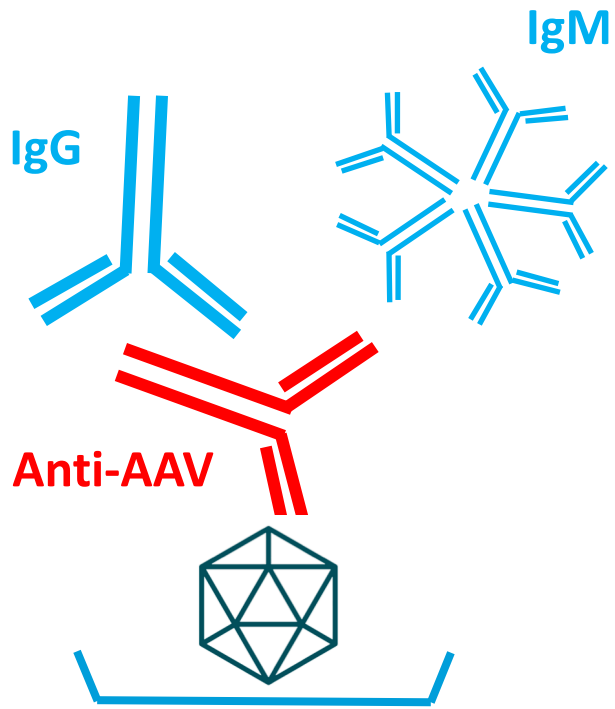
- Typically follow the 3 Tiers testing scheme
- Where there is a high incidence of AAV-positive individuals, some proceed directly to Endpoint Titer
- Additional characterizations - mostly as a legacy practice
  - Isotyping - IgG, IgM
  - Subclasses IgG1 -> IgG4
- Earlier implementation of neutralizing assays
- Neutralizing Ab assay
  - Screen and Titer
- ELISPOT cellular assessment
  - PBMCs stimulated with AAV

Bioanalytical strategies for testing – Considerations specific to Gene Therapy

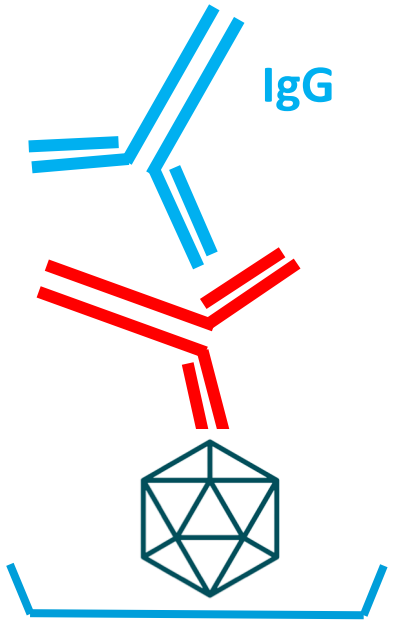
- Screening and nAb assays are likely to have significant numbers of baseline positives
  - Will require far more individuals than typical for cut-point setting
  - True for both non-clinical and clinical studies
  - May be difficult in rare disease populations, pediatric populations to obtain sufficient individuals and sample volumes
  - Significant geographic differences observed
- Implementation of cell based assays earlier than typically seen for biotherapeutics
  - Transduction-based neutralization measures
  - Determination of cellular immunity to AAV as inclusion/exclusion criteria
- Consider all foreign proteins that may be introduced/produced !
- Risk assessment early is essential if only to get started on the different assay that may be needed

Gene Therapy - Assay format – Total Antibody

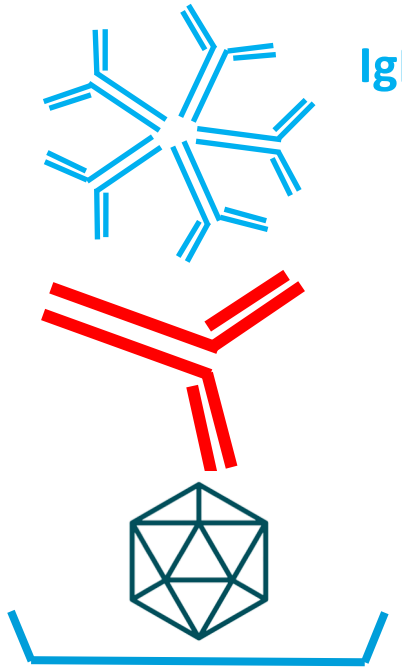
Direct Format



mixture of Anti-AAV IgGs and IgMs



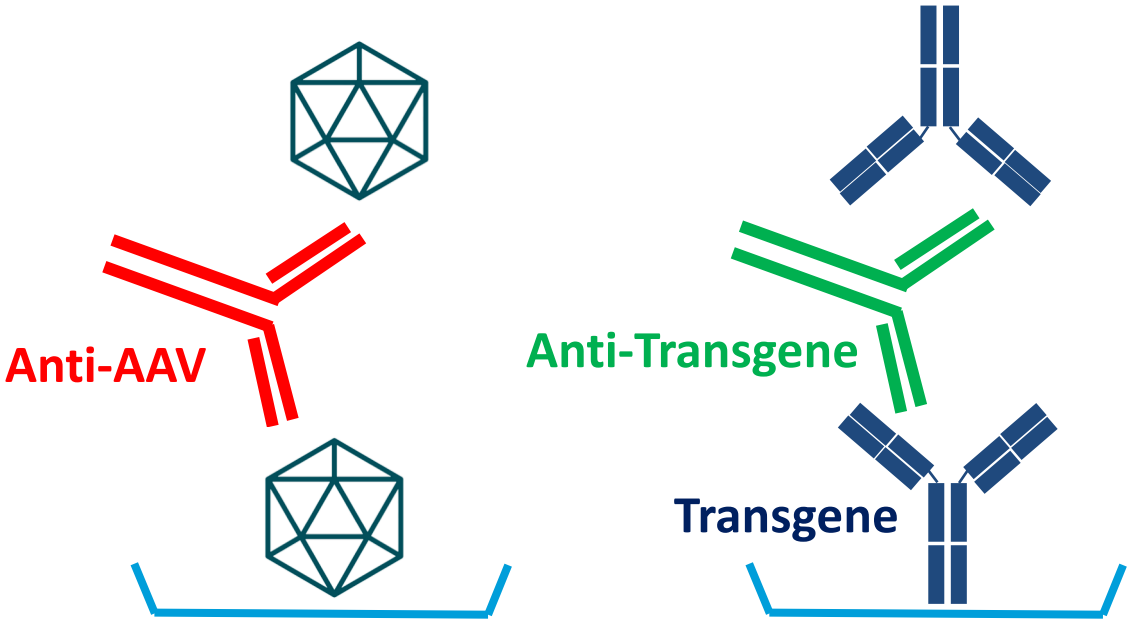
Anti-AAV IgG antibodies



Anti-AAV IgM antibodies

Gene Therapy - Assay format – Total Antibody

Bridging format



- Positive Control ?
- Use of commercial vector ?
- Use of additional assays
  - Vector
  - Transgene
- Pre-existing antibodies (to AAV, in particular) may require a larger than usual number of individuals needed for assay validation
  - ADA and nAb assay cut-point setting

Conclusions

- As therapeutic modalities increase in complexity, so too do the measures needed to quantitate and characterize them
- These new challenges also provide exciting new opportunities to set the proper precedent for measures that add scientific value
- Think carefully about what should be measured - not what can be measured
- Integrate bioanalytical data into the larger picture - not in isolation
- Regulatory guidelines are not established in many cases

Let science drives the process

Questions?

감사합니다

Thank You!

Danke

Merci

Tak!

Grazie

obrigado

ありがとう

Спасибо

Gracias

Efharisto

Tack

Contact details

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