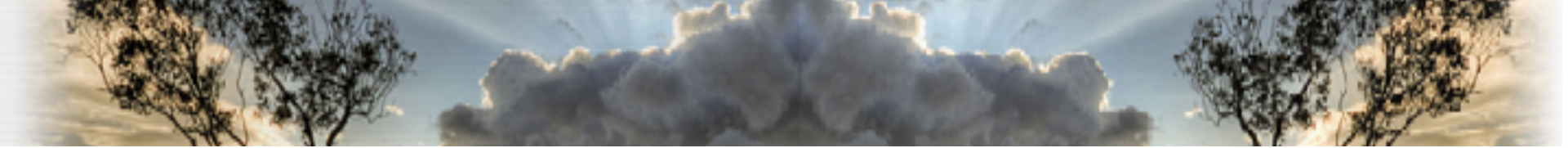

# EBF survey on regulatory challenges for Bioanalysis in C&GT

**Bioanalytical Strategies for Cell & Gene Therapies**
**EBF Training Day**

**Johannes Stanta, Chris Cox**

# Aim of session and discussion points

- Mini-Survey results from EBF Members and non-Members in C&GT
- Summary of day to day challenges
- Conclusion

# Key BioA challenges for regulatory fillings?

- Lack of guidelines on PCR assays and different interpretation of what validation means
- Shift of analytical platforms for PD Endpoints, connection between activity and concentration (e.g. for coagulation factors, enzymes)
- Immunogenicity
  - Lack of C&GT specific guideline
  - Academia based research community often not familiar with BioA's experiences in biologics development
  - Limited connectivity between clinical laboratory and BioA community. Several of the immunogenicity related assays are destined to transition into a regulated diagnostic test but there is a lack of understanding for each other's regulations and requirements (including between FDAs' departments)

# Key BioA challenges for regulatory fillings?

- Lack of consistency
  - Number of assays needed
  - Level  of assay development and qualification/validation
  - Reagent and reference standard needs
- Trying to follow guidance that have been written for biologics and is not entirely relevant for cell and viral gene therapies

# Key BioA challenges for regulatory fillings?

- Variation in level of scientific knowledge of reviewers.  Box-checkers vs understanding of the science.
- Variation in local guidance that need to be addressed. For example:
  - Brazil: provide a full statistical justification for use of non-linear calibration curve
  - China: repeat PK calculations in front of auditors

# Typical questions from regulators during Pre-filing

- Requests about immunogenicity and its assessment strategy:
  - Impact and relevance of immune responses
  - Understanding quality attributes and host cell proteins related safety risks
- Requests for assay validation data relevant to data interpretation/dose selection/patient treatment
- Need and timing for
  - total antibody assay
  - Nab assays (for patient inclusion)
  - ELISPOT assay and its data interpretation
  - End point (PD) assay quality and data interpretation

# What are the typical questions you get from regulators AFTER filing

- Questions regarding assay suitability and validation parameters
- For IND filing in immunotoxicology: always concerns if pre-clinical models are actually predictive of clinical outcomes. Hence, increasing demands for Human *in-vitro* activities

General:

- Requests for additional chromatograms and justifications supporting repeat analyses
- Cross-lab assay comparison data
- From Health Canada, verification that stability assessments were conducted in multiple tubes

# Do you feel regulator's questions are relevant to the bioanalytical challenges?

*"Depends on bioanalytical knowledge of the reviewer, some questions are check box exercises and not relevant to the C&GT, such as transgene expression that is localized to the tissue or specific site and cannot be compared to drug development of a recombinant protein"*

*"Questions that address BioA guideline are relevant. Those related to the transition of a BioA method to a diagnostic test can be viewed as BioA …or not"*

# Is there risk for a scope creep from existing BA guidance not fitting the scientific challenges for C&GT?

Guidance are written for currently established drug analyses (e.g. PK and Immunogenicity) and are in the current form inappropriate for C&GT's Mode of Action, drug, patient population and what is feasible to do.

Totally agree with this statement. Existing BioA guidance simply does not match the challenges of C&GT. I feel there is a need to address specific needs of C&GT rather than traditional areas of BioA support.

# Additional comments

- Regulators are starting to pay attention with large / mid size pharma entering the C&GT space but guidelines are still a few years away.
- Guidance is not specific to C&GT BioA. Companies are nonetheless requesting and/or following validation guidance for MAbs, Biomarkers, ADA and associated company policies and SOPs.
- Assay life cycle management — difficulties associated with long-term gene therapy follow up. What are the best practices for bridging in new reagents and QCs. How to manage the continuity of operational conditions for up to 15 years follow up?

# Day to day challenges for C&GT BioA: Anti-Vector Assays

- Use of anti-Vector antibody tests as inclusion/exclusion criterion:

Per FDA Guidance “Considerations for the Design of Early-Phase Clinical Trials of Cellular and Gene Therapy Products” assays may fall under companion or *in-vitro* diagnostic assay regulation

Outside the capability of a Bioanalysis lab, unless you work in a **medical laboratory** accredited lab?

# Day to day challenges for C&GT BioA: Anti-Vector Assays

- Anti-vector assay validation
  - blank Negative Control often difficult to find (AAV, PEG-modifications)
  - lack of guidance on cut-point evaluation, including high pre-existing antibodies
  - Vector concentration is not provided in the same way as protein concentration
    → lot-to-lot variation affects assay performance

- Irrelevant Anti-AAV assay requests:
  - CSF assays
  - Isotyping

# Day-to-day challenges: Enzymatic Assays

- Surrogate PK assay for many gene therapies
- Reference material is a surrogate or endogenous protein (not a quantitative reference standard) = all results relative
- Relative activity is more important than absolute concentration
- Enzyme calibrators – activity set for each lot, equipment, and lab
- QC values may be based on a larger dataset than traditional PK
- Higher imprecision and accuracy drift are normal (not an indication of poor assay performance)
- Life cycle management – QC limits and performance evaluated regularly

# Day-to-day challenges: Oligonucleotides

- Follow BMV PK guidance for validation
- Tissue handling/processing important for correlation with plasma PK to guide dose concentration and intervals (kinetics).
- Very sensitive assays needed
- LC-MS:
  - Lack of stable internal standards
  - Nonspecific binding
  - Adduct ion formation causes reduction in sensitivity
  - Evaluate carryover
  - Short column life span
- LBA (hybridization assays):
  - New lab skills, reagents and equipment needed

# Day-to-day challenges: Flow cytometry and cellular kinetics

- No official validation guidelines. Many follow CLSI:H62 and white papers
- Complex technology with no reference material, cross-instrument standardization
- Matrix and stability is a challenge
- Very different considerations for
  - Accuracy
  - Linearity
  - Selectivity
  - Prozone
  - Quantitation range
  - No ISR

# Conclusions

1. Questions from regulators are mostly fair
2. Start the interaction with regulators early
3. Industry and regulators need to step out of comfort zone of existing regulations
4. There is a need to bring together departments with traditionally different backgrounds (immunology, diagnostic, biomarker, BioA)
5. Share and develop industry best practice

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Thank You !

# Contact Information

Questions: info@e-b-f.eu

**European Bioanalysis Forum vzw**

www.e-b-f.eu