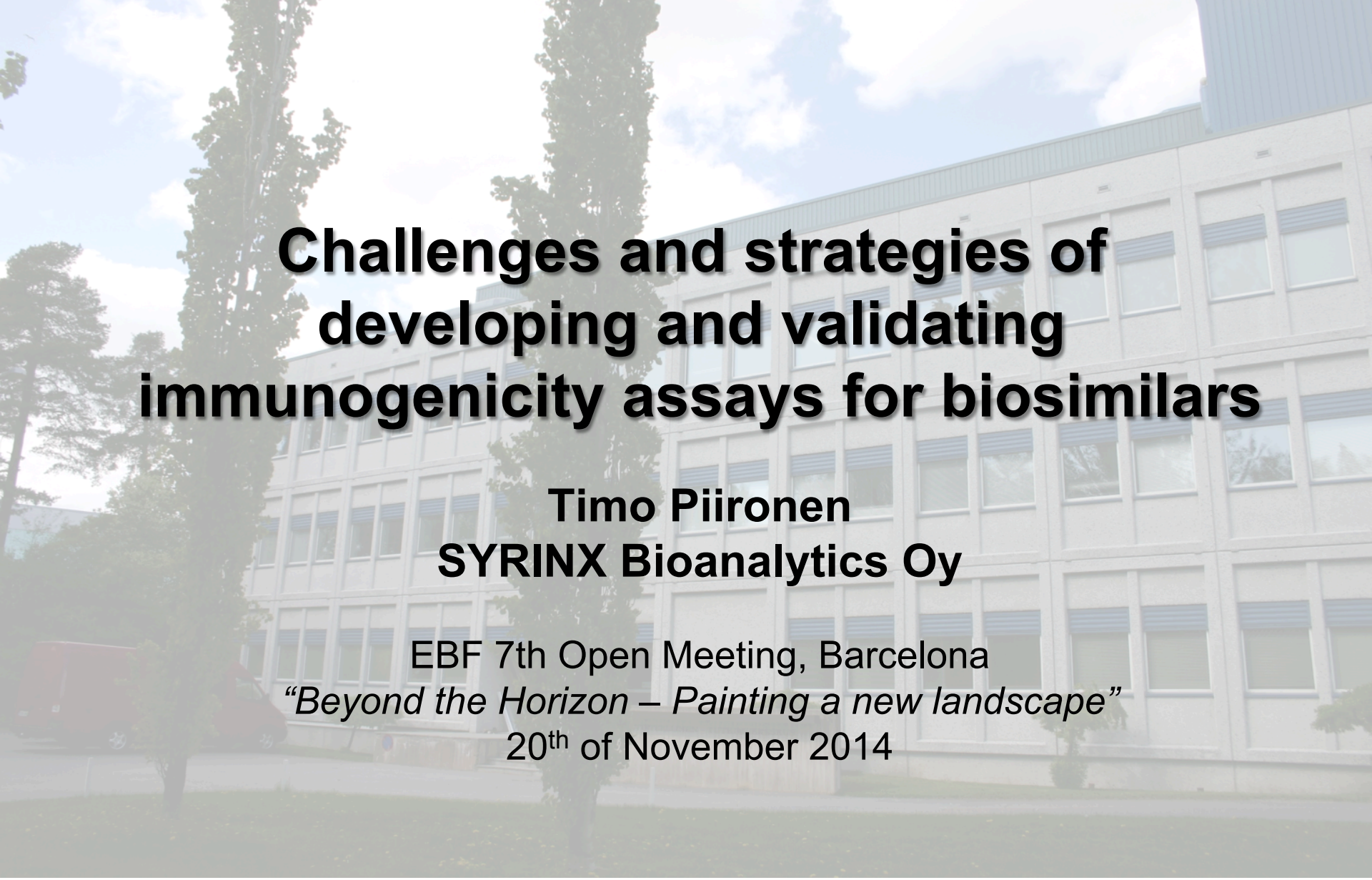


Challenges and strategies of developing and validating immunogenicity assays for biosimilars

Timo Piironen
SYRINX Bioanalytics Oy

EBF 7th Open Meeting, Barcelona
"Beyond the Horizon – Painting a new landscape"
20th of November 2014

SYRINX BIOANALYTICS

Session program

16:15	18:00	Biosimilars
16:15	16:35	**Timo Piironen (Syrinx Bioanalytics)** Challenges and strategies of developing and validating immunogenicity assays for biosimilars
16:35	16:55	**James Munday (Covance)** The use of PK, PD and ADA bioanalysis for evaluation of the overall Immunogenicity of biosimilars and the bioanalytical challenges for determining if there are equivalent safety risks.
16:55	17:15	**Ricardo Gutierrez-Gallego (Anapharm Biotech)** Biosimilarity assessment - impact on safety and efficacy
17:15	17:30	**Joseph C. Marini (Janssen R&D, on behalf of AAPS LBABFG)** Recommendations from the AAPS LBABFG Biosimilars Action Program Committee on the Development and Validation of PK and ADA assays for Biosimilar Drug Development
17:30	17:45	**Birgitte Buur Rasmussen (Ferring Pharmaceuticals, on behalf of EBF)** Recommendations from EBF biosimilars evaluation group
17:45	18:00	**Panel Discussion**

Contents (focus on bioanalytical issues)

- Definitions and background
- Pros and cons of 1-assay vs. 2-assay strategies (ADA screening assays)
- Development and validation of immunogenicity assays (1-assay strategy)
- Practical considerations
- Conclusions

Biosimilars

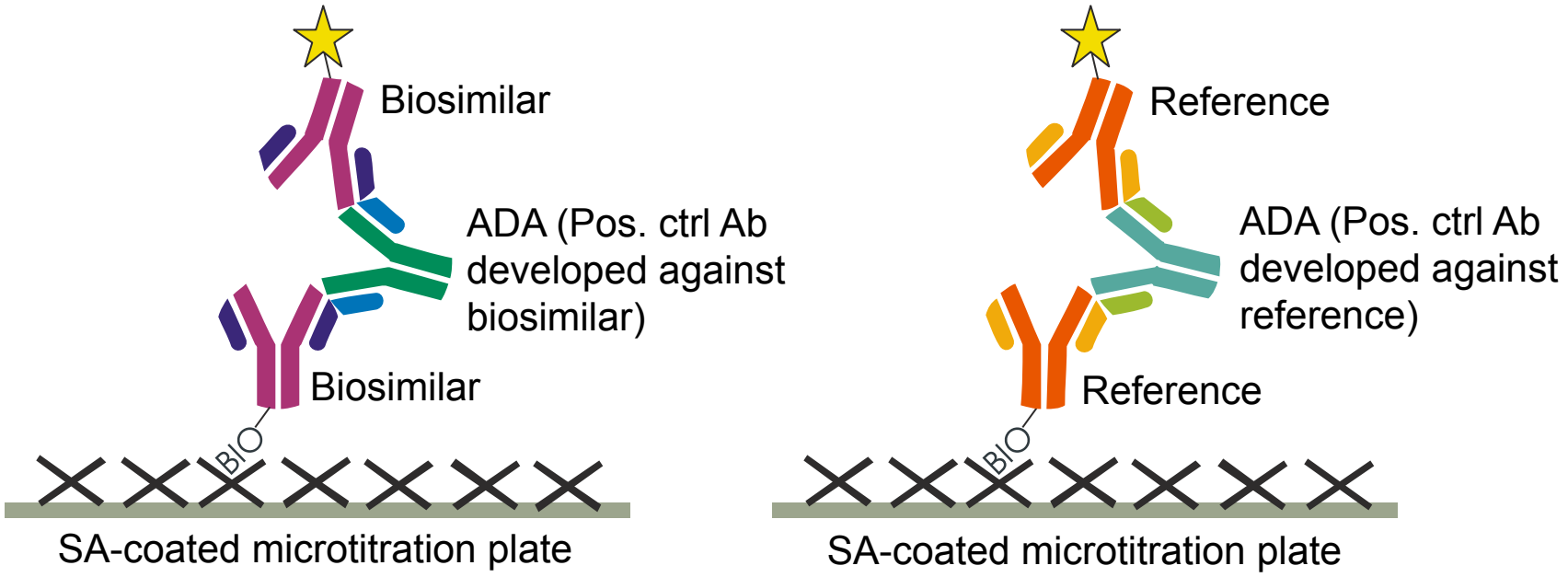
- **Biosimilar** (**follow-on biopharmaceutical**) is a medicine whose active substance is similar to that of a biological medicine that has been already authorized (innovator/ originator/reference product).

Background

- Many patents for innovator products will expire in the near future.
- Biological products from different sources cannot be assumed to be bioequivalent or have the same immunogenicity profile even if identical genes are expressed in the same host cells due to various small modifications of the products (biosimilars are not generics).
- Properties of biologicals which may affect immunogenicity are also dependent on many factors such as downstream processing and formulation (e.g. impurities, infectious agents and aggregation).
- Most critical safety concern is immunogenicity.
- It will be critical to demonstrate the similarity of the biosimilar compared to the innovator/reference product → increased immunogenicity with biosimilar?

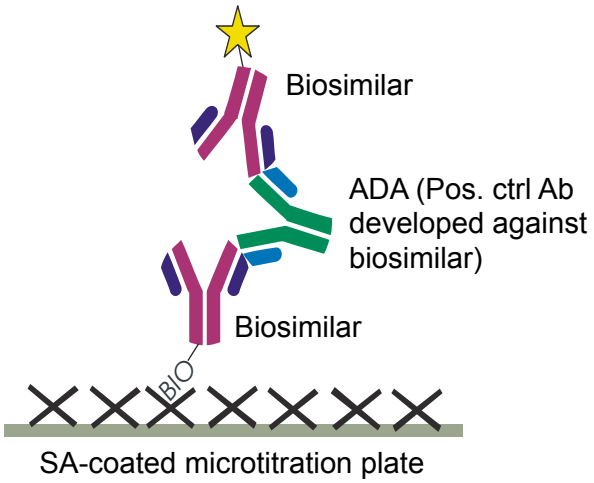
One or two ADA assays?

Two assays

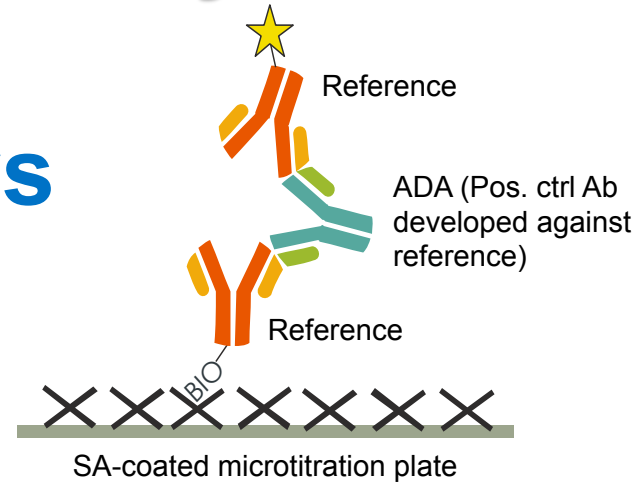


Both assays should be comparable with their own cut point, confirmation assay cut point, precision, sensitivity and drug tolerance.

One or two ADA assays?



Two assays



Pros:

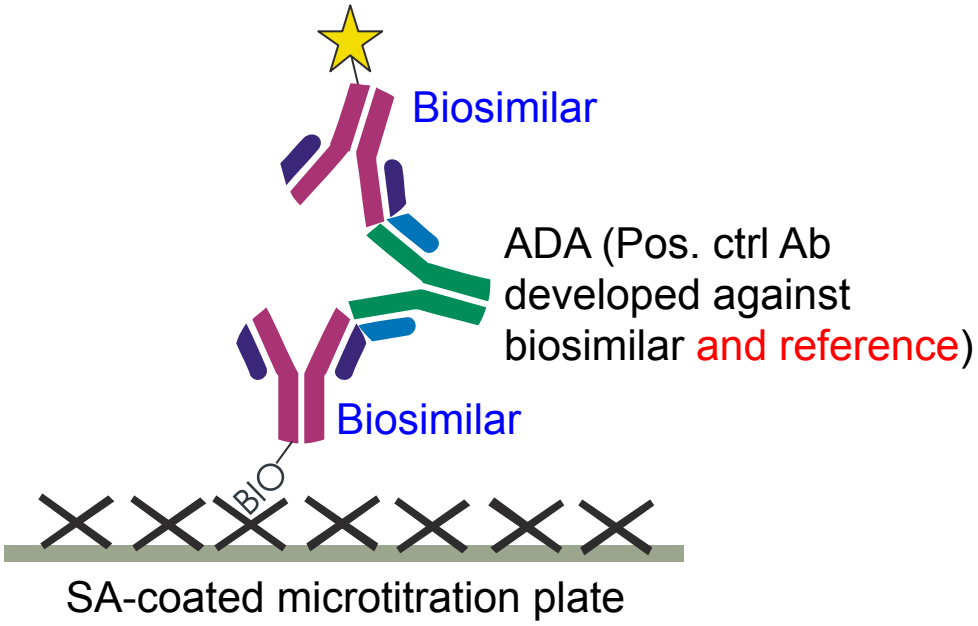
- All possible differences in immunogenicity can be detected between biosimilar and reference products

Cons:

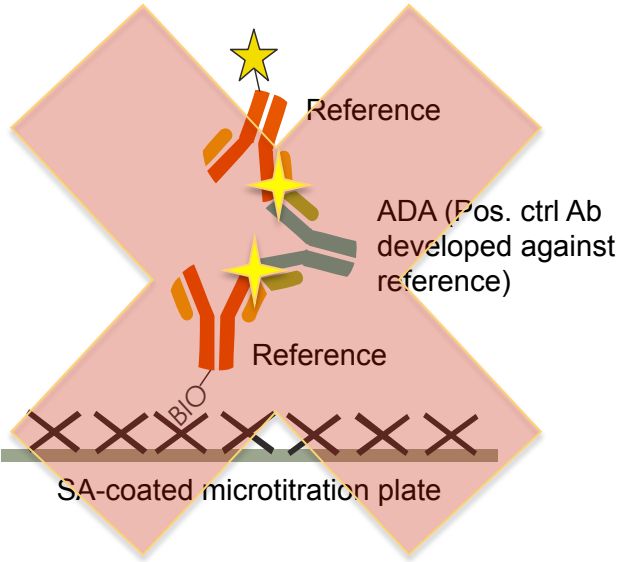
- Both assays have to be comparable, i.e. they have to have same sensitivity and precision (very difficult to achieve and show in practise with two sets of reagents, cut points etc.)
- Additional sample volume needed in blinded study setting

One or two ADA assays?

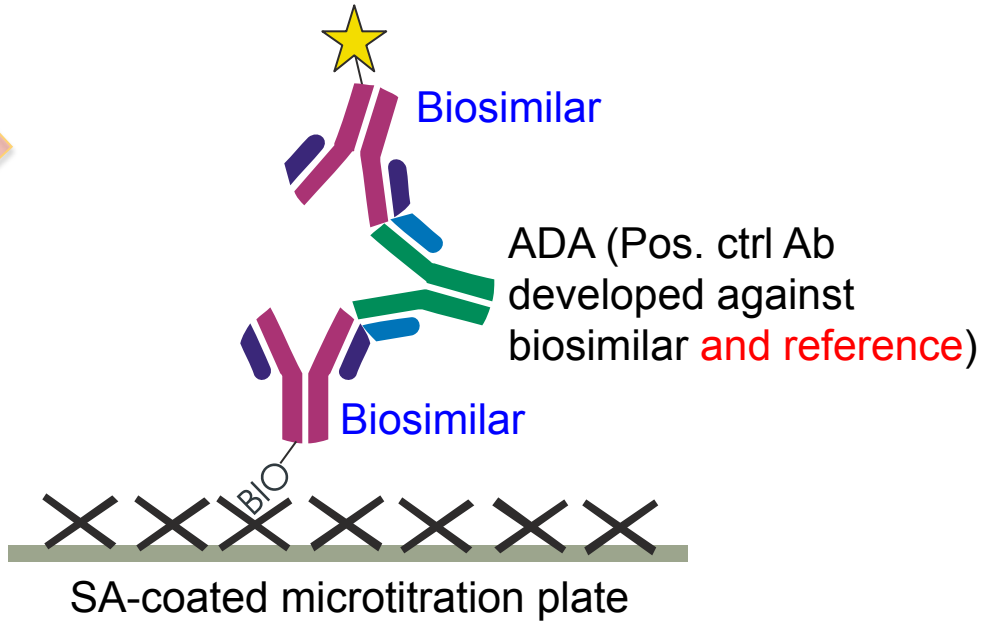
One assay



One or two ADA assays?



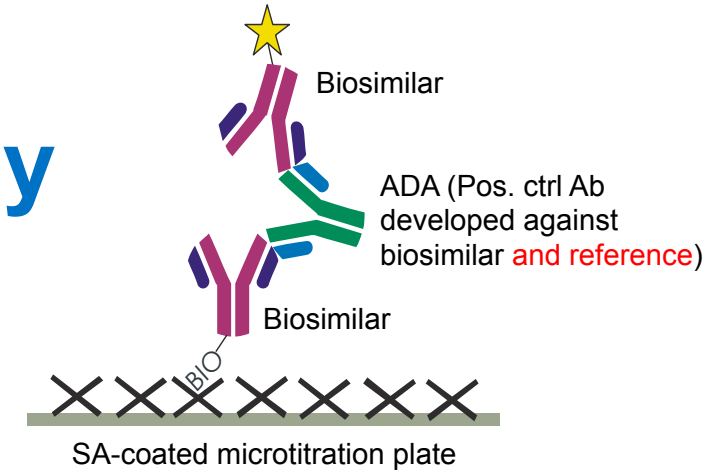
One assay



Can be justified as it results in optimal detection of biosimilar ADA!

One or two ADA assays?

One assay



Pros:

- Less variation due to the use of one assay and one set of reagents
- Method validation / in-study QCs done with only one assay and pos. ctrl Ab, e.g. sensitivity, drug tolerance and cut point
- No difficulties in blinded stydy setting (lower sample concumption)
- Saves time and money

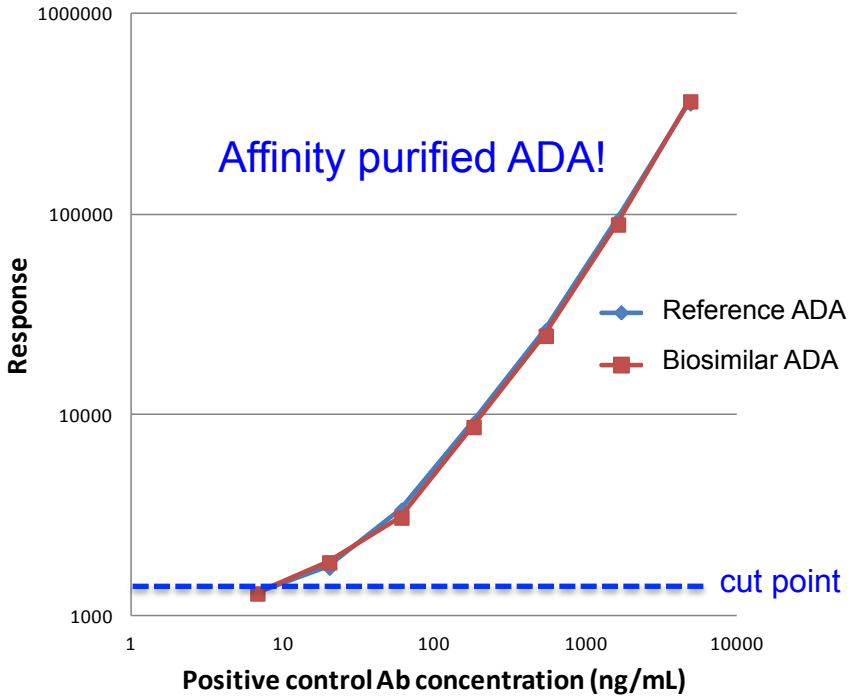
Cons:

- ADA developed against innovator structures not present in biosimilar molecule not detected

Development of ADA assays

Aim to develop one assay, then need to check following:

- Sensitivity: Dose-response curve comparison with pos. ctrl Abs against biosimilar and reference (comparable assay sensitivity?)



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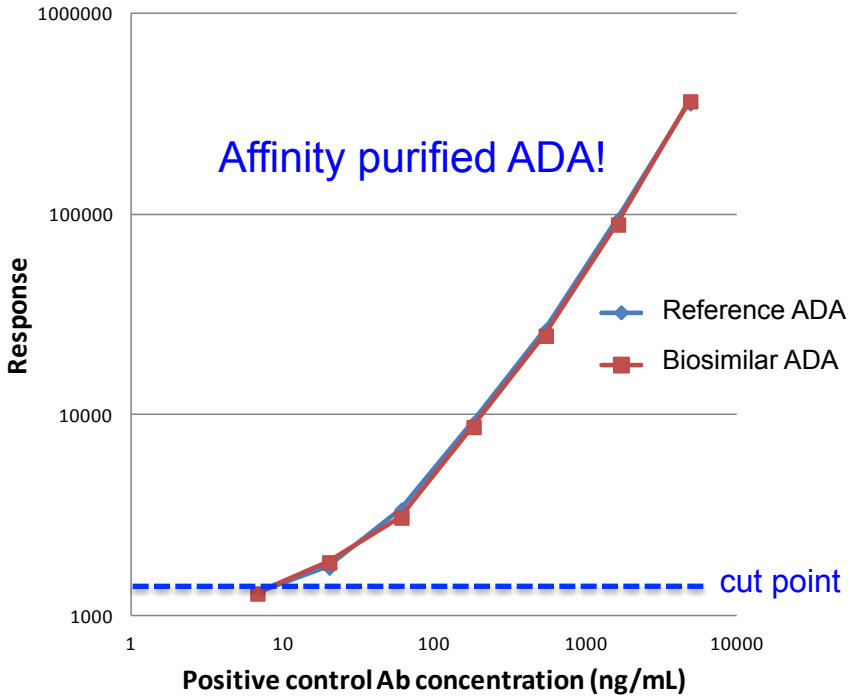
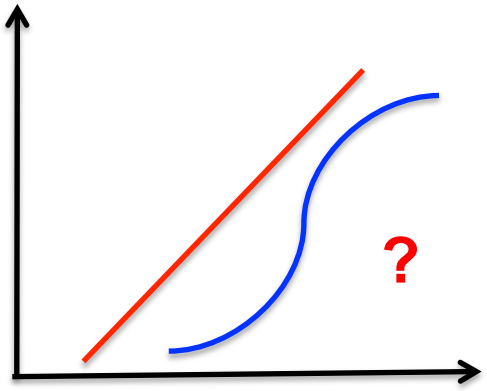


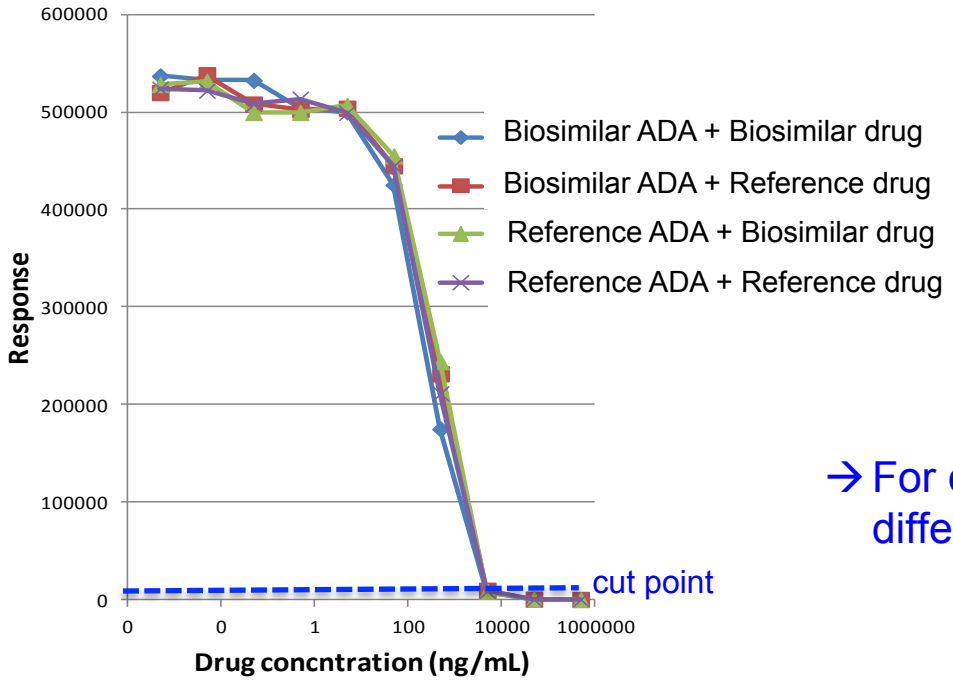


→ However, different individual animals may produce different antibody isotypes and affinities!
→ For qualitative assays 3-fold differences are acceptable?

Development of ADA assays

Aim to develop one assay, then need to check following:

- Drug tolerance: Pos. ctrl Abs against biosimilar and reference inhibited with biosimilar and reference drugs (cross-check)



→ For qualitative assays 3-fold differences are acceptable?

Validation of ADA assays (1-assay)

- Validation parameters, such as cut point, confirmation assay cut point, sensitivity, selectivity, precision, drug tolerance recovery and stability, are performed with biosimilar reagents only.
- Test drug tolerance with biosimilar ADA (low and high pos. ctrl), depleted with biosimilar and reference drugs?

  → Acceptance criteria: For qualitative assays 3-fold differences are acceptable?

  → Use only biosimilar ADA throughout the study for depletion in the confirmation assay?

Development and validation of NAB assays

- Development and validation of NAB assays is somewhat similar to the ADA screening assay strategy:
  - 1- or 2-assay?
  - If 1-assay strategy is selected, focus on investigating selectivity (titration of cell based assay signal), sensitivity and drug tolerance during method development with biosimilar and reference drug/ADA.

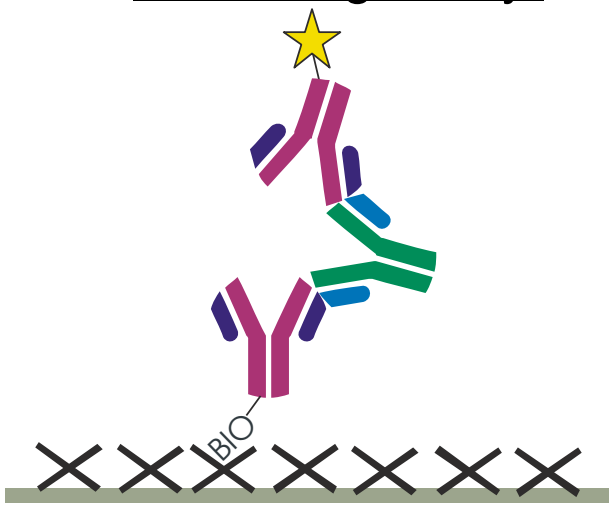
Practical considerations

- **Availability of anti-drug control antibodies**
  - Commercial sources (reference ADA)?
  - Own antibody production (biosimilar and reference ADA)?
- **Selection of ADA screening assay format**
  - Selection case-by-case based on required sensitivity, drug tolerance etc. (does not have to be the same format as was used originally with the reference drug)
- **Selection of NAB assay format**
  - Selection case-by-case based on required sensitivity, drug tolerance etc. (does not have to be the same format as was used originally with the reference drug)
- **Confirmation assay?**
  - Use only biosimilar drug for depletion for all study samples?
- **Immunogenicity rate may be difficult to measure particularly at low incidence?**

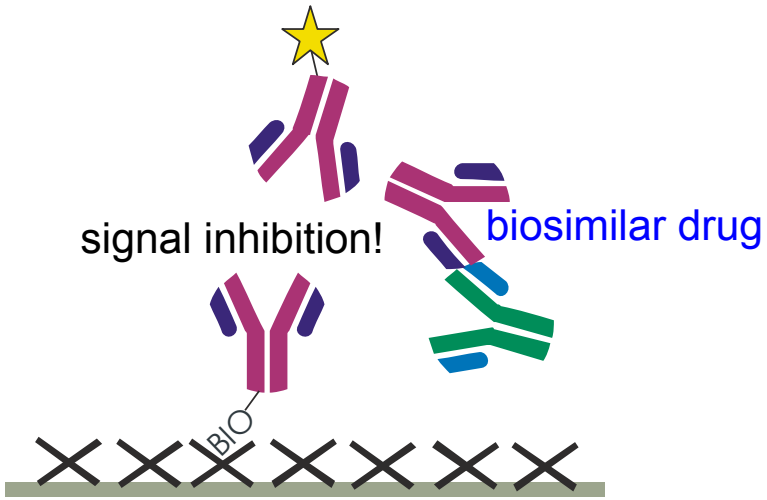
Conclusions

- **In most cases using one assay screening ADA strategy is adequate:**
  - **One pos. ctrl Ab (biosimilar ADA) and one confirmation assay reagent (biosimilar drug)**
  - **Details about acceptance criteria needs to be discussed**

Screening assay:



Confirmation assay:



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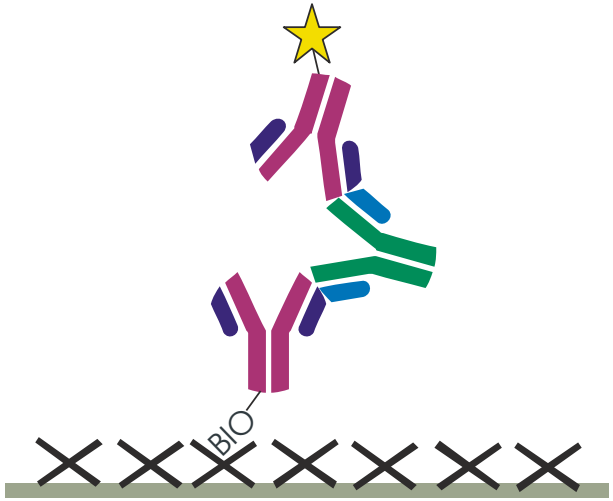
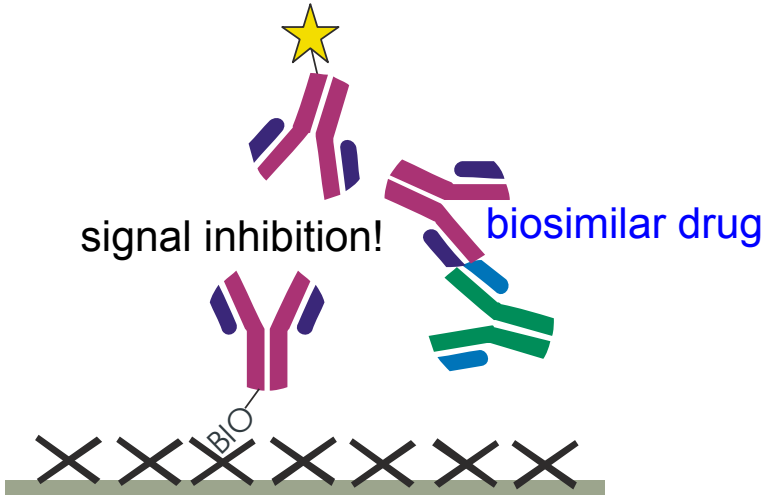


Confirmation assay:



However, case-by-case evaluation may result in selecting the 2-assay approach!

Thank you!

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