Clinical ADA Monitoring in High-Dose Biologics

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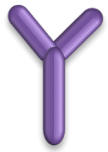
NOVARTIS | Reimagining Medicine

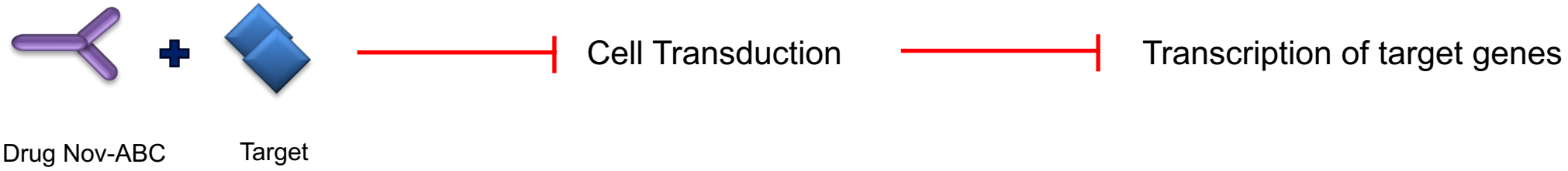
Nov-ABC, a drug administered at high-dose with high frequency



Drug Nov-ABC

Humanized IgG mAb

Mechanism of Action



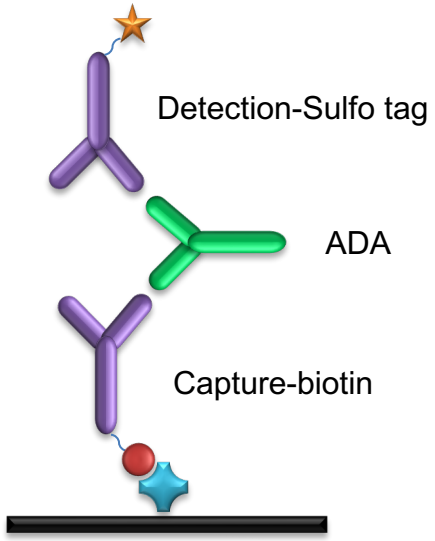
Clinical study	Study design	Dosing RDE	Ctrough	Target (total accumulation)
Phase 1	Dose escalation + dose expansion	20-30 mg/kg, Q3W	500 – 800 µg/mL	25X that of baseline
Phase 2	Safety + Randomized part	30 mg/kg, Q3W or Q2W	500 – 800 µg/mL	

Drug Interference

Target Interference

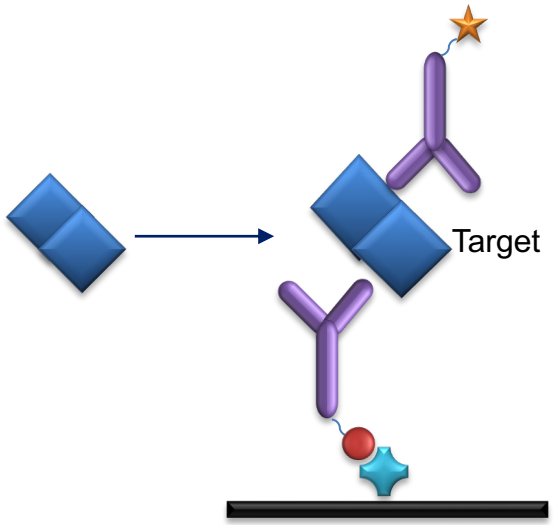
Anti-Nov-ABC ADA assay: interference from target and drug

ADA detection (no interference)



Homogenous bridging assay using chemiluminescence, a standard approach for ADA detection

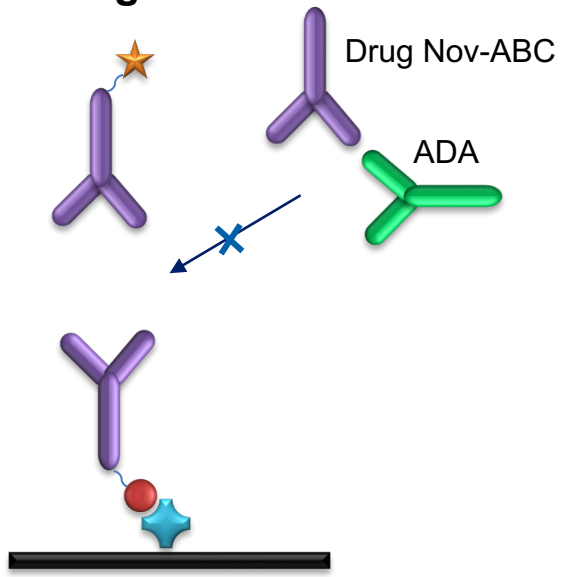
Target interference



Target is detected by the assay

consequence = false positives

Drug interference

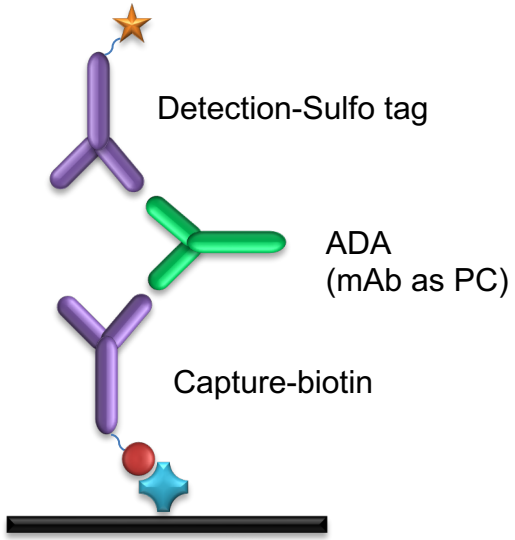


In the presence of high concentrations of Novartis-ABC drug, majority of ADA are bound to drug

consequence = false negatives

Classical acid dissociation could not be implemented in the assay due to the biology of the target

Anti-Nov-ABC ECLIA ADA method: assay performance



Validation Parameter		ADA Assay (in healthy matrix)
Minimal required dilution (MRD)		1:50
Sensitivity		50.0 ng/mL of PC in 100% serum
Hook effect		No hook effect up to 58.0 µg/mL of drug
Screening	Cut-point factor (CPF)	1.17
Confirmatory	Confirmatory assay cut-point (CCP)	22%
Titer	Titer cut-point (TCP)	1.17

Drug interference	
	Drug Nov-ABC (µg/mL)
ADA (ng/mL) - Positive Control (PC)	100
mAb clone (origin: mouse hybridoma)	**<0.0781**

Target interference, RLU (PC= mAb clone)		
	Drug Nov-ABC (µg/mL)	
	0	100
Target (pg/mL)	Response (ECL)	
0 to 1000	-	-

Strong drug interference

Using the mAb clone, the assay has apparent low drug tolerance

No target interference

Target interference tested up to 1000 pg/mL

Strategies tested for improvement of DT

- **ECLIA without acid dissociation**:
  - Tested drug tolerance at sensitivity levels higher than 100 ng/mL (250, 500 ng/mL & above) relative to surrogate antibody (mAb)
  - Anticipated drug exposure 100-1000 µg/mL

- **Testing of four alternative sample pretreatments**:

Antibody competition	⊘ increased interference and noise
ACE: Affinity capture elution	⊘ increased target interference due to acid treatment
PANDA: Precipitation and Acid dissociation	⊘ increased target interference due to acid treatment
SPEAD: Solid-Phase Extraction with Acid Dissociation	⊘ increased target interference and noise

- **For upcoming clinical studies**
  Based on its overall performance, bridging ECLIA (without acid dissociation) was selected for full validation

Relevance of drug tolerance (DT) assessments

- Sensitivity and DT depend on choice of positive control antibody
  consider binding affinity, epitope specificity, etc.

- According to FDA guidance (2019):
  *"The sponsor may examine drug tolerance by deliberately adding **different known amounts of positive control** antibody into ADA-negative control samples in the absence or presence of different quantities of the therapeutic protein product to determine whether the therapeutic protein product interferes with ADA detection."*

  ➢ **Characterization of DT at different levels of PC required**

- Recommended sensitivity by FDA guidance (2019): 100 ng/mL
  Might not always be clinically relevant, e.g. 100 ng/mL of ADA* in presence of 800 µg/mL drug mAb → effective concentration of drug = 799.9 µg/mL

- Therefore:
  - ➢ The ECLIA ADA assay described before represented a good compromise between drug tolerance (DT), target interference (TI), shows low background & adequate sensitivity
  - ➢ Based on the overall performance of the assay, the project team recommended to move forward with sample analysis of Phase 1 clinical study
  - ➢ **In parallel, interaction with FDA initiated to discuss the overall ADA strategy**

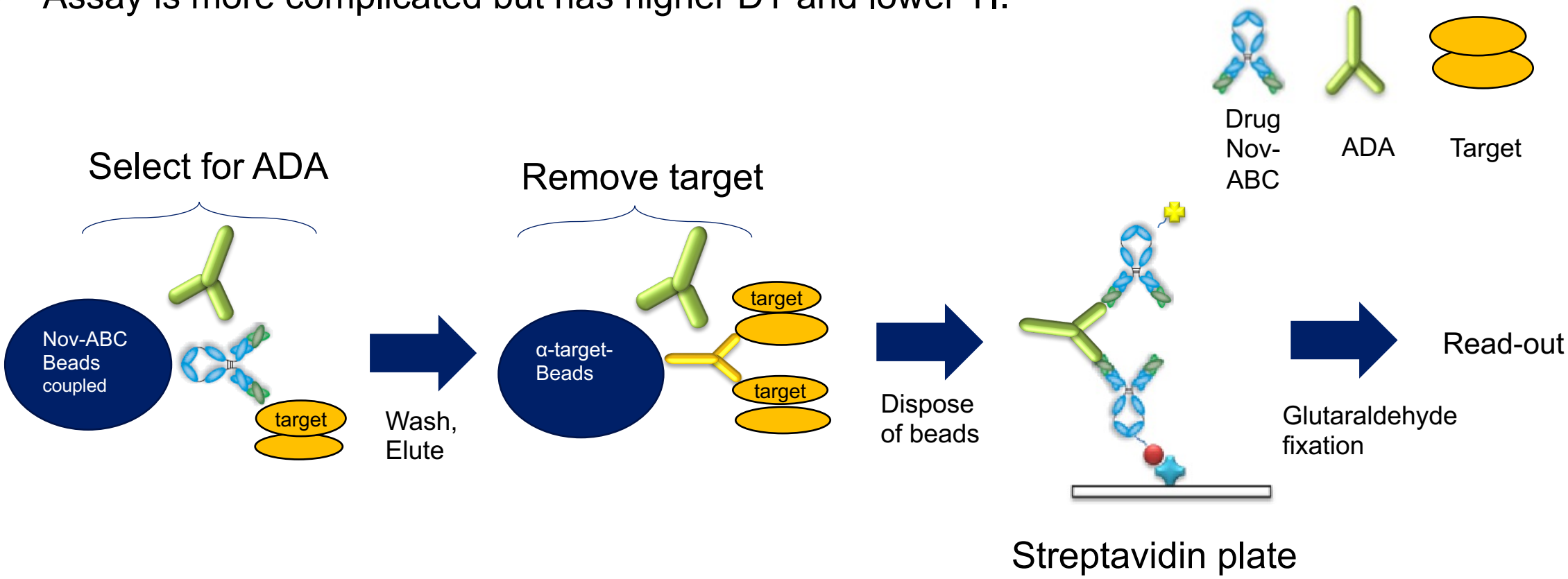
**assuming ADA is mostly IgG*

First Interaction with the FDA

- **Meeting:** Type B
- **Document:** Investigational new drug application (IND)
- **Response:**
  - Additional development of ADA assay is necessary for the concerned antibody & target to provide more meaningful results related to the ADAs development against the drug.
  - “Sufficient patient samples should be banked for analysis by this and other assays that may be developed”
  - Current bridging format would not be capable of being sufficiently sensitive that can provide meaningful results at high drug concentrations in serum other than washout phase

Anti-Nov-ABC ADA bead-based method

- NVS developed and qualified a new assay which enriches ADAs and removed target
- Assay is more complicated but has higher DT and lower TI.



*Glutaraldehyde fixation is to prevent the plate bound complexes from falling apart & degradation of the plate signals during read-out

Anti-Nov-ABC ADA bead-based method: assay performance

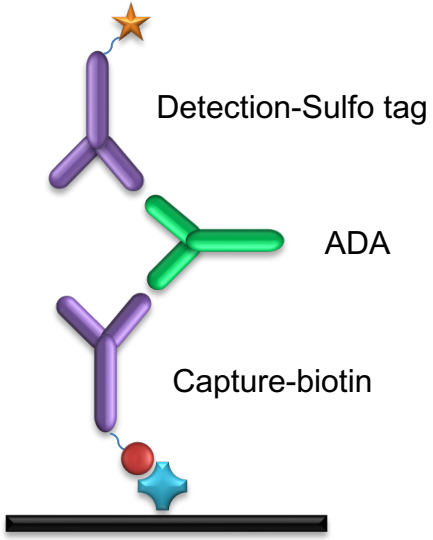
Validation Parameter		ADA Assay (in healthy matrix)
Minimal required dilution (MRD)		1:25
Sensitivity		15.0 ng/mL of PC in 100% serum
Precision (%CV)		0.6 and 141% (average: 10.0 - 127%)
Assay passing rate		47 runs out of 62 (75%)
Screening	Cut-point factor (CPF)	1.13
Confirmatory	Confirmatory assay cut-point (CCP)	31.9%
Titer	Titer cut-point (TCP)	1.28

Drug interference		
	Drug Nov-ABC (µg/mL)	
ADA (ng/mL) - Positive Control (PC)	100	5000
mAb anti-ID (origin: recombinant Ab expressed in HEK293-6E cell line)	**1000**	**1000**

Method is better but has drawbacks

Target interference (PC= mAb anti-ID)			
	0	100	600
	Drug Nov-ABC (600 µg/mL)		
Target (ng/mL)	Response (ECL)		
0	-	+	+
5	-	+	+
10	-	+	+
20	-	+	+

Anti-Nov-ABC ECLIA ADA method: DT & TI using different positive controls



Additional positive controls were introduced and characterized for DT and TI

Drug interference				
	Drug Nov-ABC (µg/mL)			
ADA (ng/mL) - Positive Control (PC)	100	500	2000	10,000
mAb clone (origin: mouse hybridoma)	<10	<10	59.9	162
mAb anti-ID (origin: mouse hybridoma)	**120**	**435**	**764**	**>1500**
pAb (origin: rabbit)	**<10**	**80.5**	**358**	**1283**

Target interference, RLU (PC= mAb anti-ID)				
	Drug Nov-ABC (µg/mL)			
	0	100	500	1000
Target (ng/mL)	Response (ECL)			
0	-	-	-	-
2	+	-	-	-
5	+	+	-	-
20	+	+	-	-

Second Interaction with FDA

- **Meeting:** Type D
  - New Type D meeting authorized in 2022 under PDUFA VII, providing the opportunity to address a narrow set of issues on a shorter timeline than other meeting types
  - Limited to no more than 2 focused topics
  - Meeting package (briefing book) must be submitted at the same time as meeting request
  - FDA response expected within 50 days of receipt of request (vs. 75 days for Type C meetings)
  - Written response only (WRO) requested in this case

Metrics and Outcome of the Type D meeting

For Type D meetings, the Agency must receive the background information from Sponsor at the time of the meeting request.

Metrics:

- Time elapsed between our decision to contact FDA and Briefing book (BB) completion → 30 WD
- Briefing book → 12 pages (1 question + background information)
- Time elapsed between reception of BB by FDA and their response = 34 WD

Outcome:

- Additional DT study data provided for positive control (PC) antibodies show enhanced drug tolerance compared to original PC used during method validation study and supports the continued development of the ECLIA method.
- Prior to patient sample analysis:
  - Cut-point and use of normalization factor should be assessed and submitted for review
  - Target interference in presence of multiple positive controls should be assessed

Conclusions & Summary

- Although our assay was suitable to support clinical studies, but characterization of our assay with only 1 PC at 1 concentration level did not indicate on suitable assay performance
- Following FDA feedback, we initiated development of additional ADA assay → more sensitive, improved drug tolerance and target interference
- Newly developed bead-based ADA assay improved drug tolerance and sensitivity, however the assay lacked substantial improvement in terms of robustness or precision
- Multiple methods were tested, but none improved drug tolerance without impacting target interference
- Additional positive controls were introduced & characterized further for drug tolerance & target interference
- Second interaction with FDA resulted in agreement of the fully validated ECLIA ADA assay as showing enhanced drug tolerance and continued method development. However, prior to SA, cut-point should be determined from drug-naïve patient population and use of normalization or cut-point factor should be assessed

Key take-away messages

Our learning and recommendations what to do when you have high doses:

- Characterize the assay as much as you can with multiple positive controls (mAb & pAb) & at different concentration levels of PC
- Invest in the production of multiple tools

We were successful in engaging with FDA in a recently implemented Type D meeting to align on clear path forward for clinical ADA assay strategy

Engage in cross-functional collaboration with Clin Pharm, RA and Medical team

Bring BA question to health authority in context with clinical relevance, safety & dose regimen

Thank you.

And to all the co-authors and collaborators:

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