



# Towards assessment of clinically relevant immunogenicity of monoclonal antibodies

Theo Rispens

## Why monitor ADA?

- Potential issues
  - loss of efficacy (which will be related to PK)
  - adverse effects
- Why measure ADA at all?
  - PK / target binding of drug can be assessed directly
  - Both loss of response and adverse effects: clinical assessment
- Reasons to measure ADA:
  - Identify reason for loss of response / adverse event
  - Prediction of loss of response / adverse events



# Outline

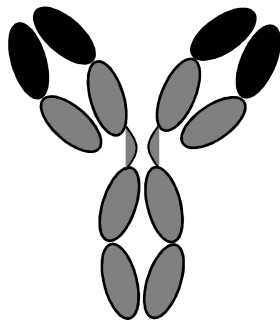
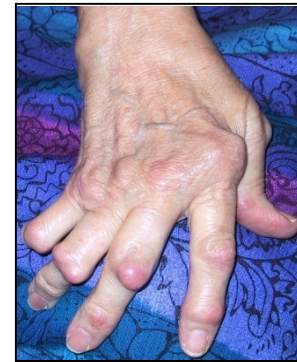
- ADA and clinical response
- ADA and adverse events
- Transient responses & early prediction

# TNF blockers

Inflammation in e.g.

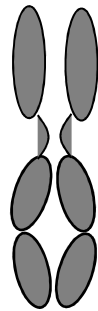
- Rheumatoid arthritis
- Crohn's disease
- Psoriasis

can be suppressed by blocking TNF:



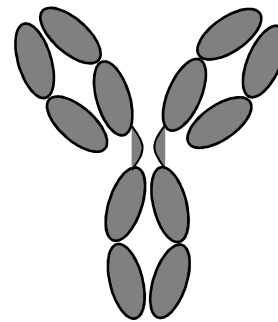
Infliximab

1998



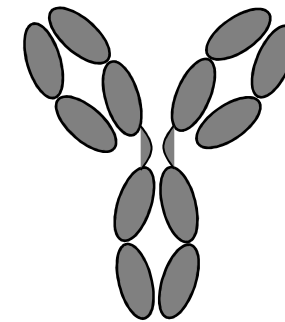
Etanercept

1998



Adalimumab

2002



Golimumab

2008



Certoluzimab

2009



# Therapeutic drug monitoring and immunogenicity testing – Amsterdam area

- systematic collection of data and serum samples of patients treated with biologicals
- inflammatory conditions: rheumatoid arthritis, psoriatic arthritis, psoriasis; multiple sclerosis, ...



Academisch Medisch Centrum  
Universiteit van Amsterdam





## Relation between ADA and clinical response

# Development of Antidrug Antibodies Against Adalimumab and Association With Disease Activity and Treatment Failure During Long-term Follow-up

British Journal of Dermatology (2015) 173, pp855–8.

The correlation of clinical efficacy, serum trough levels and antidrug antibodies in ustekinumab-treated patients with psoriasis in a clinical-practice setting

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Lucien Aarden, PhD

Gerrit Jan Wolbink, MD,

JAMA. 2011;305(14):1460-1468

**Context** Short-term data on the immunogenicity of monoclonal antibodies showed associations between the development of antidrug antibodies and diminished serum drug levels, and a diminished treatment response. Little is known about the clinical relevance of antidrug antibodies against these drugs during long-term follow-up.

**Objective** To examine the course of antidrug antibody formation against fully hu-

## Development of Antiinfluximab Antibodies and Relationship Clinical Response in Patients With Rheumatoid Arthritis

ARTHRITIS & RHEUMATISM Vol. 54, No. 3, March 2006, pp 711–715

Gerrit Jan Wolbink,<sup>1</sup> Marijn Vis,<sup>2</sup> Willem Lems,<sup>2</sup> Alexandre E. Voskuyl,<sup>3</sup> Els de Groot,<sup>4</sup> Michael T. Nurmohamed,<sup>5</sup> Steven Stapel,<sup>4</sup> Paul P. Tak,<sup>6</sup> Lucien Aarden,<sup>4</sup> and Ben Dijkmans<sup>3</sup>

### Clinical relevance: concentration and anti-natalizumab antibodies in multiple sclerosis

MULTI SCLER JOURN EXTENDED REPORT Kneepkens EL, et al. *Ann Rheum Dis* 2013;

### Immunogenicity, adalimumab levels and clinical response in ankylosing spondylitis patients during 24 weeks of follow-up

Anke Vennegoor<sup>1</sup>, Theo RispeMS<sup>2</sup>, Eva MM Strijbis<sup>1</sup>, Alexandra Seewann<sup>1</sup>, Bernard MJ Uitdehaag<sup>1,3</sup>, Lianne Frederik Barkhof<sup>4</sup>, Chris H Polman<sup>1</sup>, Gertjan Wolbink<sup>2</sup>, and Joep Killestein<sup>1</sup>

Eva L Kneepkens,<sup>1</sup> James Cheng-Chung Wei,<sup>2</sup> Michael T Nurmohamed,<sup>1,3</sup> Kai-Jieh Yeo,<sup>2</sup> C Y Chen,<sup>2</sup> Irene E van der Horst-Bruinsma,<sup>1,3</sup> Desiree van der Klei Theo RispeMS,<sup>5</sup> Gertjan Wolbink,<sup>1,5</sup> Charlotte L M Krieckaert<sup>1</sup>

### concentrations in psoriatic arthritis; an association with disease activity at 28 and 52 weeks of follow-up

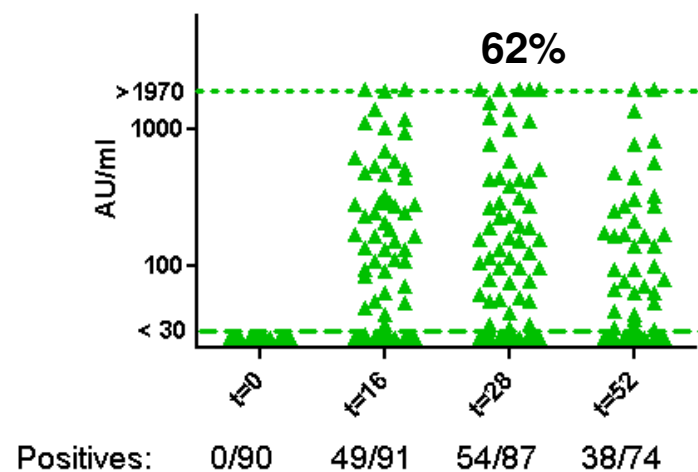
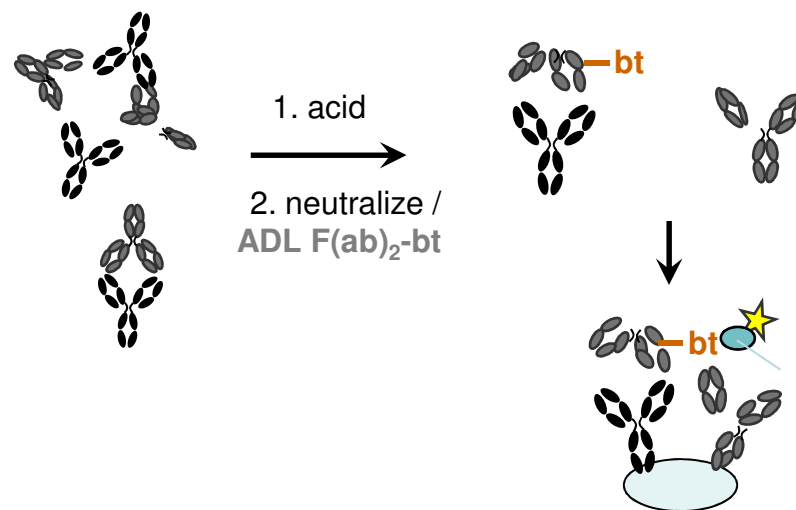
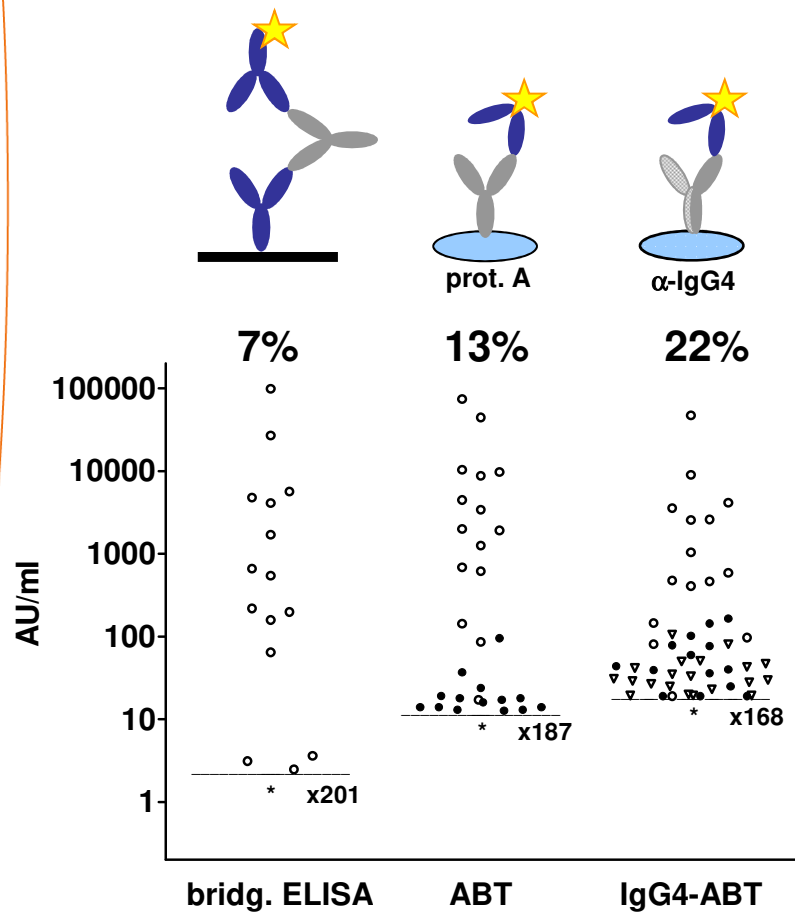
Vogelzang EH, et al. *Ann Rheum Dis* 2014,

Erik H Vogelzang,<sup>1</sup> Eva L Kneepkens,<sup>1</sup> Michael T Nurmohamed,<sup>1</sup>

Arno W R van Kuijk,<sup>1</sup> Theo RispeMS,<sup>2</sup> Gertjan Wolbink,<sup>1,2</sup> Charlotte L M Krieckaert<sup>1</sup>

## Anti-adalimumab in RA patients on standard adalimumab dose after 28 weeks of treatment

- different numbers of ADA+ patients in different assays





## Imraldi (SB5)

## Cyltezo (BI 695501)

### NHV Immunogenicity Results

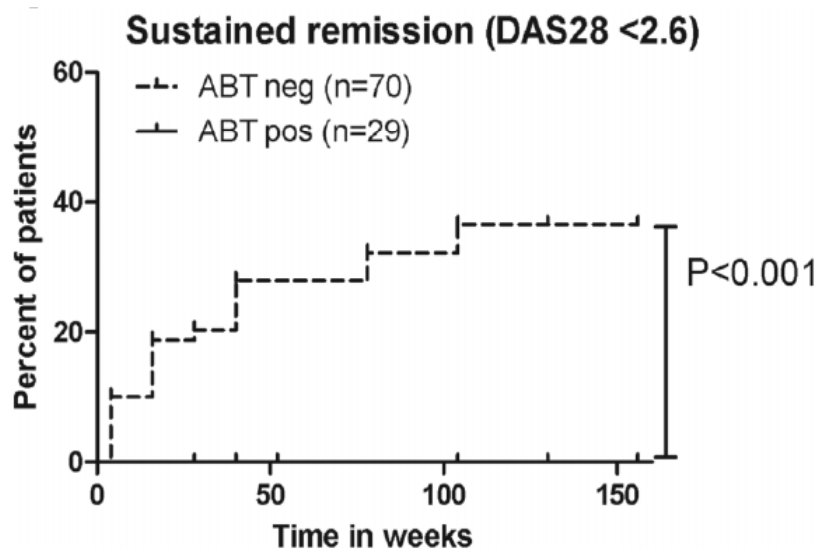
Treatment	Overall ADAs <sup>2</sup> n/n <sup>1</sup> (%)	Overall NAbs n/n <sup>1</sup> (%)
Single dose <i>s.c.</i> injection of 40 mg SB5	62/63 (98.4%)	49/62 (79.0%)
Single dose <i>s.c.</i> injection of 40 mg EU Humira <sup>®</sup>	60/63 (95.2%)	48/60 (80.0%)
Single dose <i>s.c.</i> injection of 40 mg US Humira <sup>®</sup>	63/63 (100%)	52/63 (82.5%)

**Table 9. Incidence of ADAs in Healthy Subjects, Study 1279.**

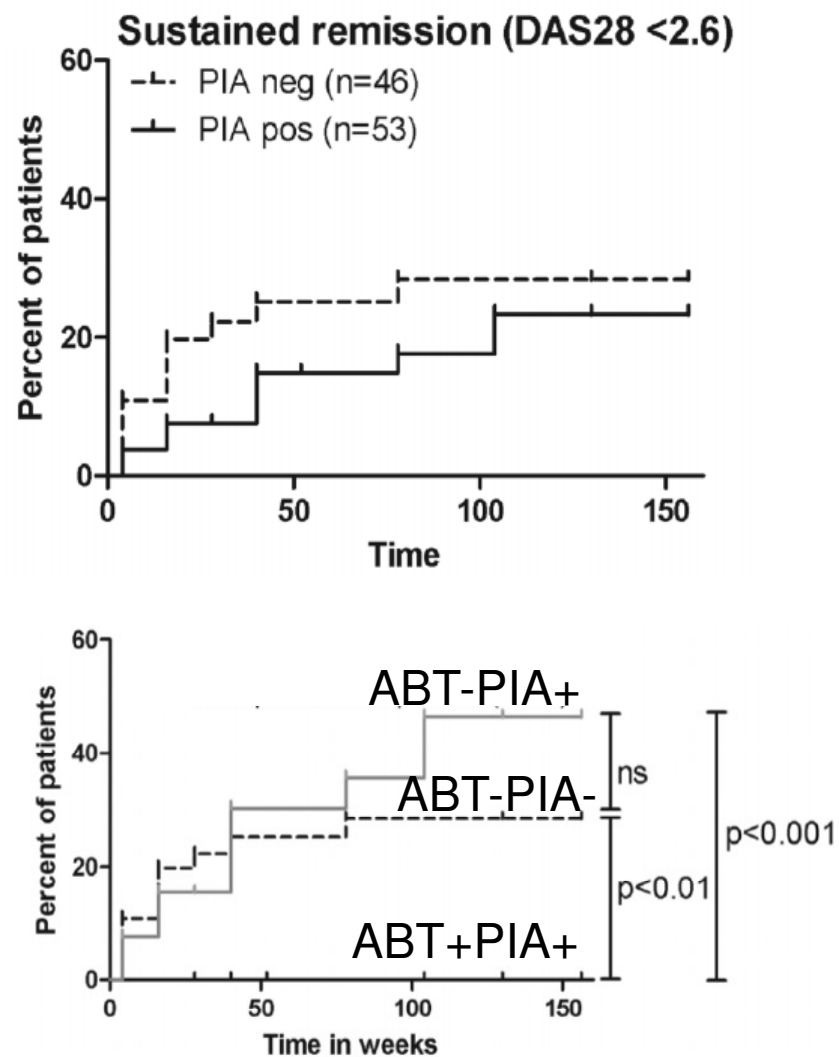
Timepoint	ADA positive		
	BI 695501 (n=108)	US-Humira (n=108)	EU-Humira (n=108)
Baseline	4 (4%)	3 (3%)	4 (4%)
Day 8	35 (33%)	6 (6%)	5 (5%)
Day 14	44 (41%)	38 (35%)	21 (20%)
Day 28	50 (47%)	60 (56%)	40 (37%)
Day 44	62 (58%)	59 (55%)	58 (54%)
Day 56	85 (80%)	82 (77%)	78 (74%)
Day 71 (EOS)	99 (93%)	95 (88%)	91 (84%)

# Correlation with clinical outcome depends on ADA assay

ABT (drug-sensitive)

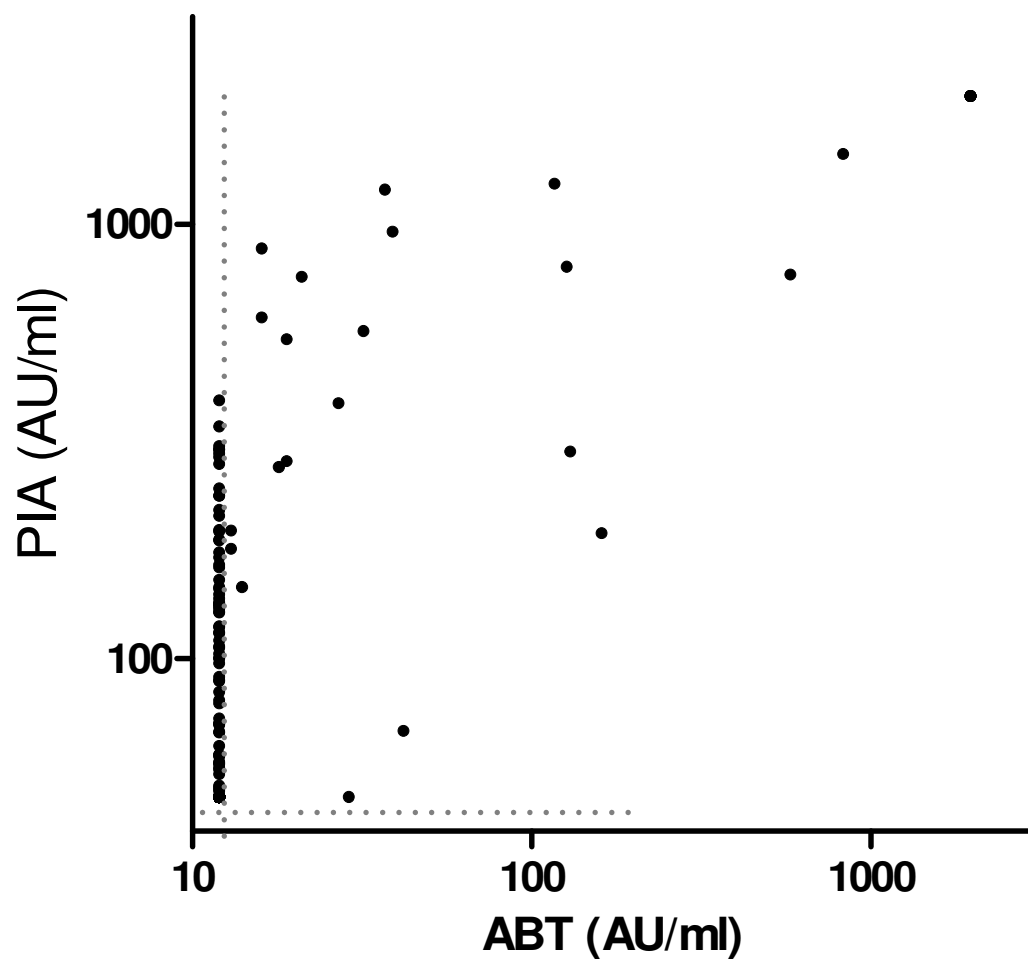


PIA (drug-tolerant)



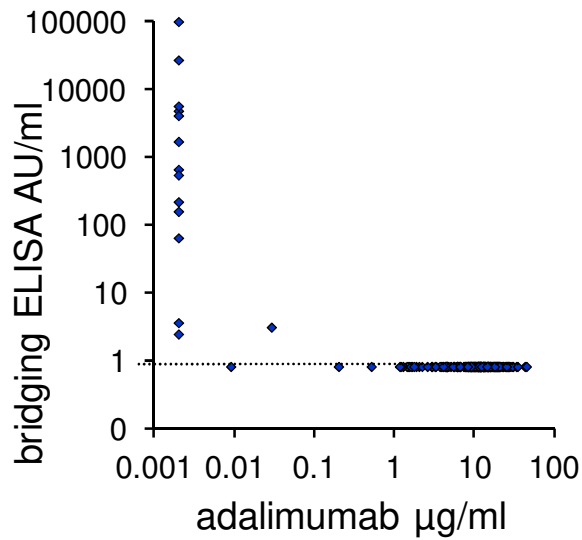
## Drug-tolerant vs drug-sensitive: (semi-)quantitative comparison

ADA titer in adalimumab-treated RA patients  
drug-tolerant (PIA) vs drug-sensitive (ABT)

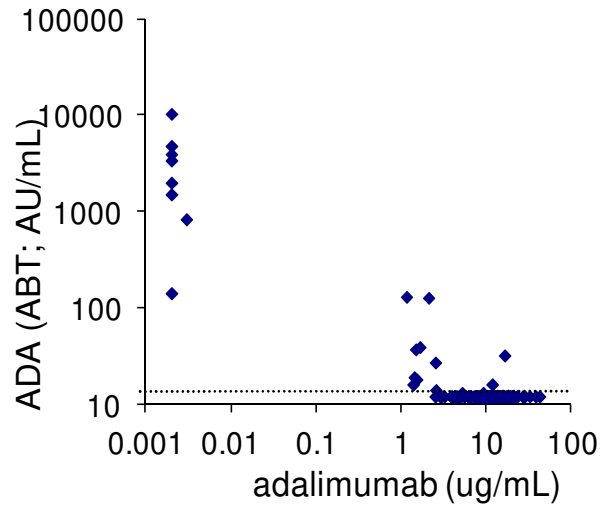


# Drug levels vs anti-drug antibodies: a balance

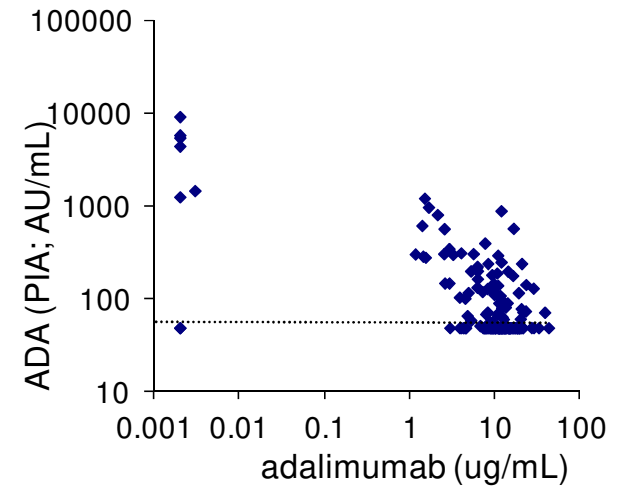
bridging elisa vs PK



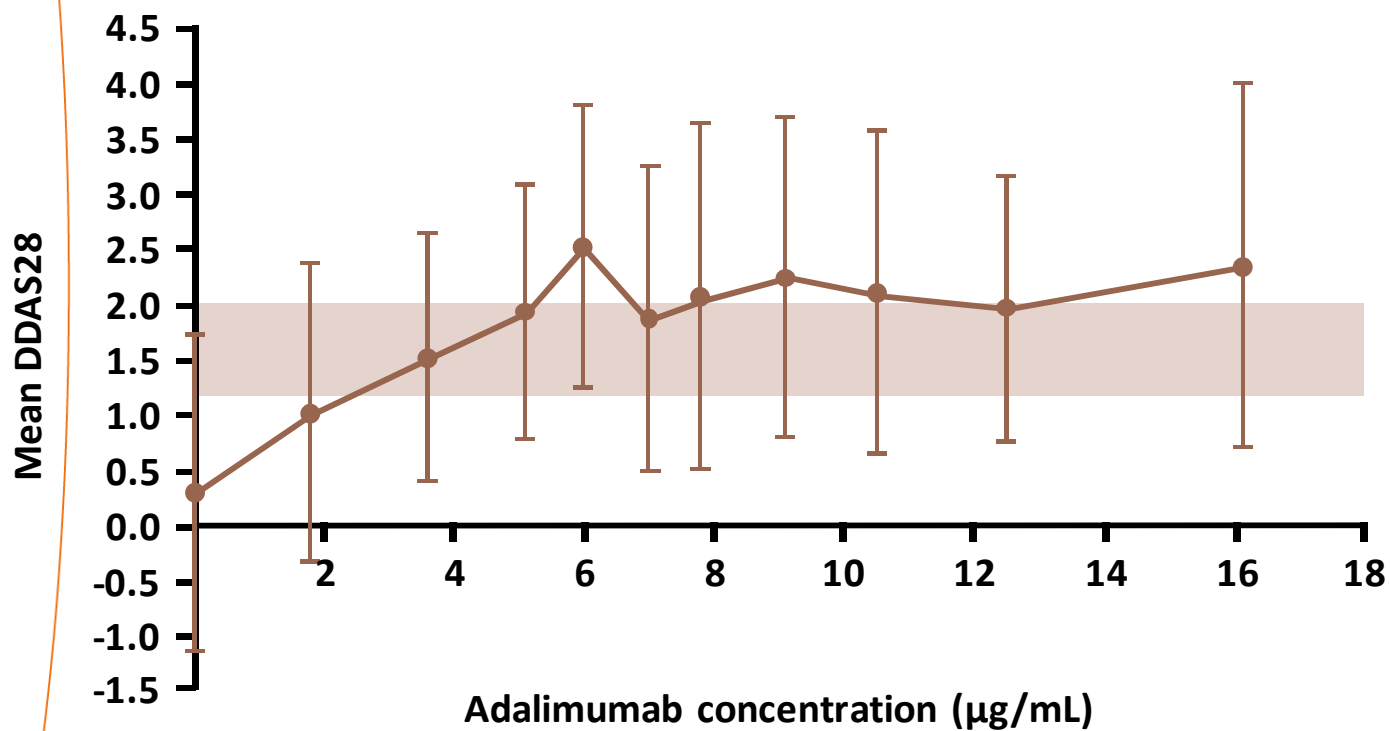
ABT vs PK



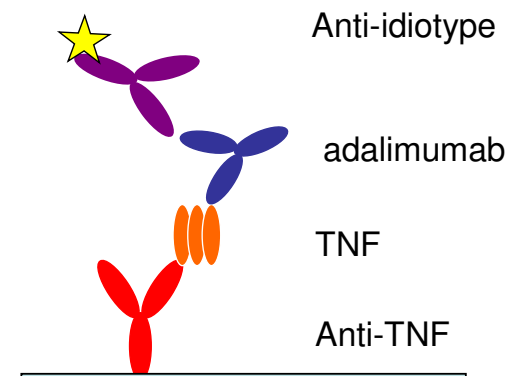
PIA vs PK



# Concentration-effect curve (adalimumab/RA)

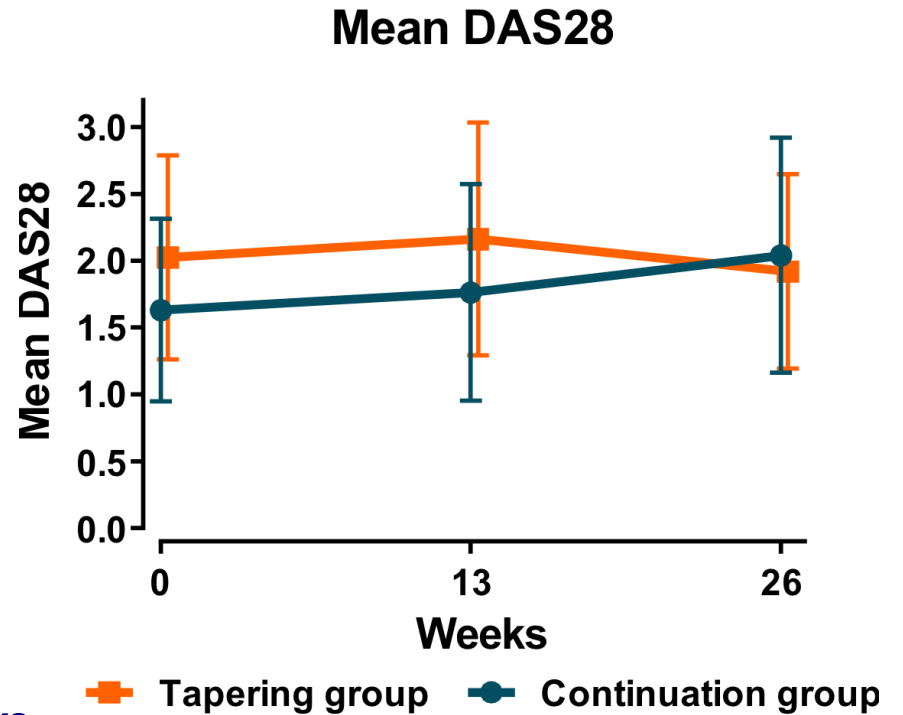
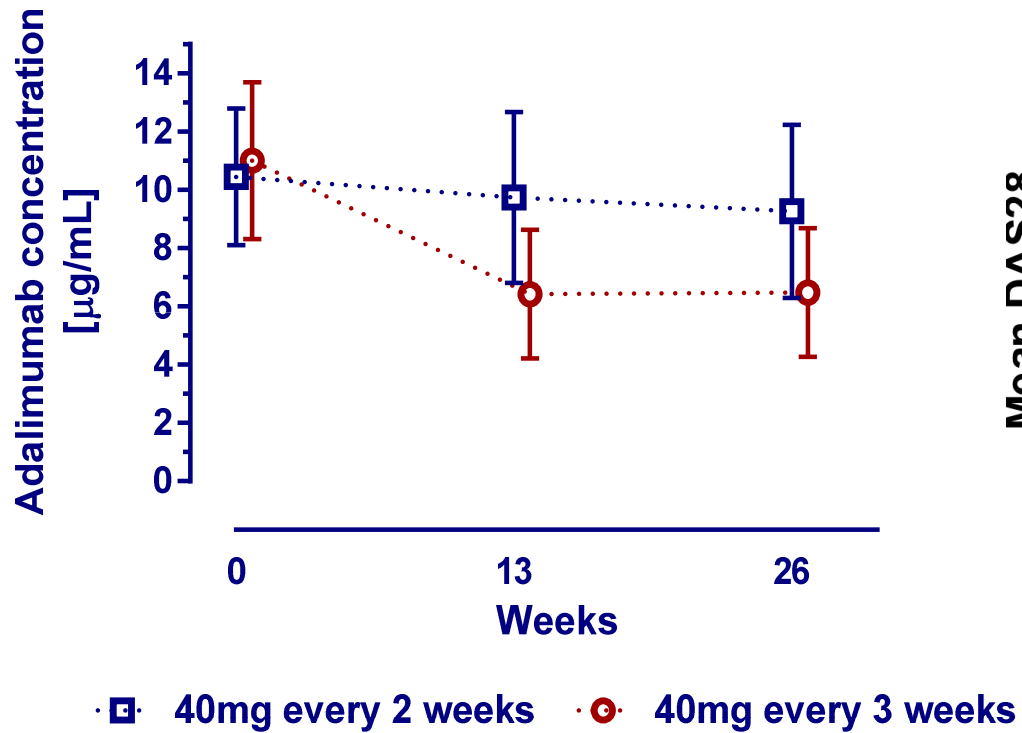


Each dot is mean of 20 patients  
through concentrations vs DDAS28 at week 28



# RA-TDM Study

## dose-reduction if adalimumab >8mg/L



## Dose reduction / treatment discontinuation (without TDM)

Disease activity guided dose reduction and withdrawal of adalimumab or etanercept compared with usual care in rheumatoid arthritis: open label, randomised controlled,

non-inferiority trial [thebmj](#) | *BMJ* 2015;350:h1389 | doi: 10.1136/bmj.h1389

Noortje van Herwaarden,<sup>1</sup> Aatke van der Maas,<sup>1</sup> Michiel J M Minten,<sup>1</sup> Frank H J van den Hoogen,<sup>1,2</sup> Wietske Kievit,<sup>3</sup> Ronald F van Vollenhoven,<sup>4</sup> Johannes W J Bijlsma,<sup>5</sup> Bart J F van den Bemt,<sup>6,7</sup> Alfons A den Broeder<sup>1</sup>

### RESULTS

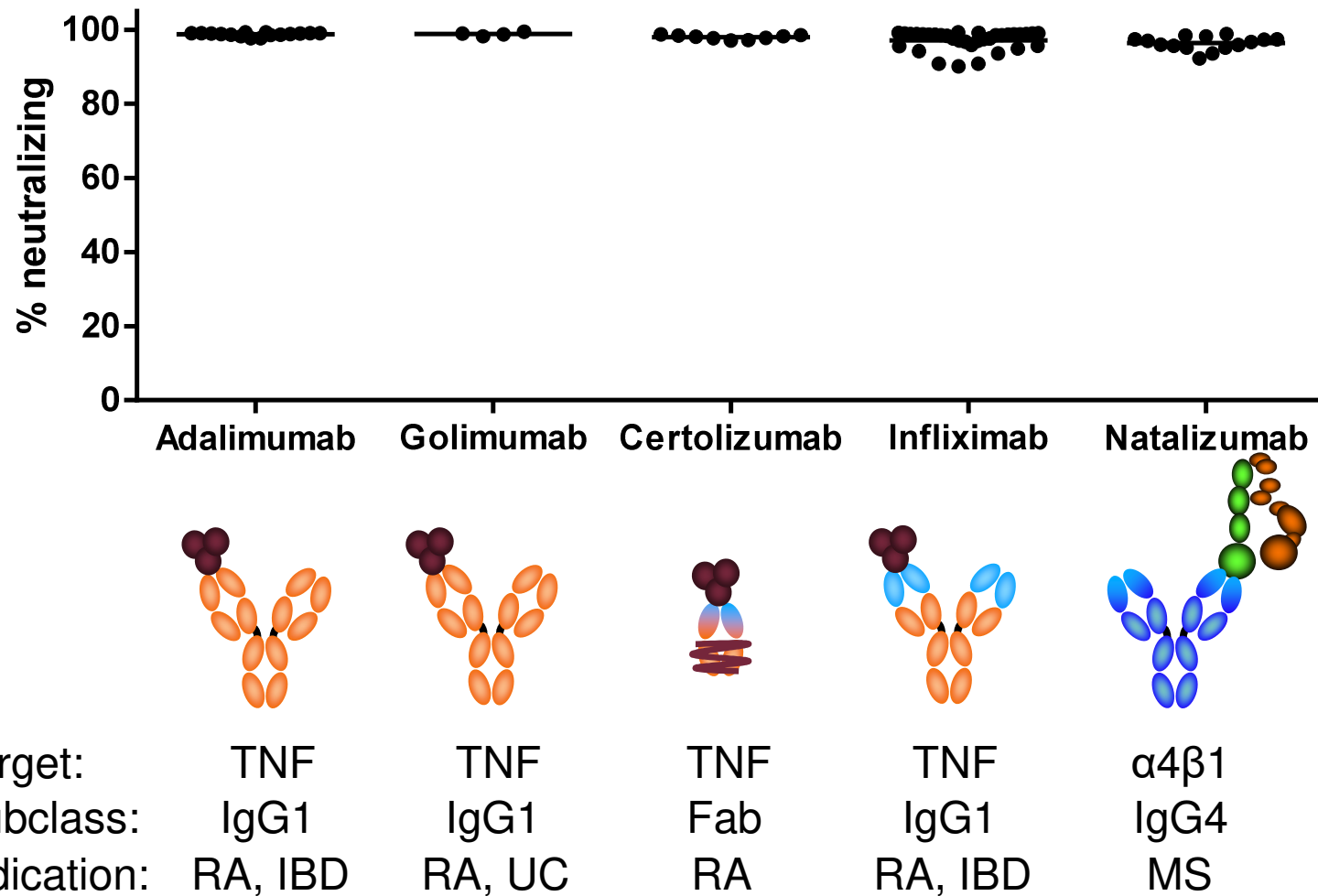
Dose reduction of adalimumab or etanercept was non-inferior to usual care (proportion of patients with major flare at 18 months, 12% v 10%; difference 2%, 95% confidence interval -12% to 12%). In the dose reduction group, TNF inhibitor use could successfully be stopped in 20% (95% confidence interval 13% to 28%), the injection interval successfully increased in 43% (34% to 53%), but no dose reduction was possible in 37% (28% to 46%). Functional status, quality of life, relevant radiographic progression, and adverse events did not differ between the groups,

although short lived flares (73% v 27%) and minimal radiographic progression (32% v 15%) were more frequent in dose reduction than usual care.

## Immune Complexes: Neutralization & clearance

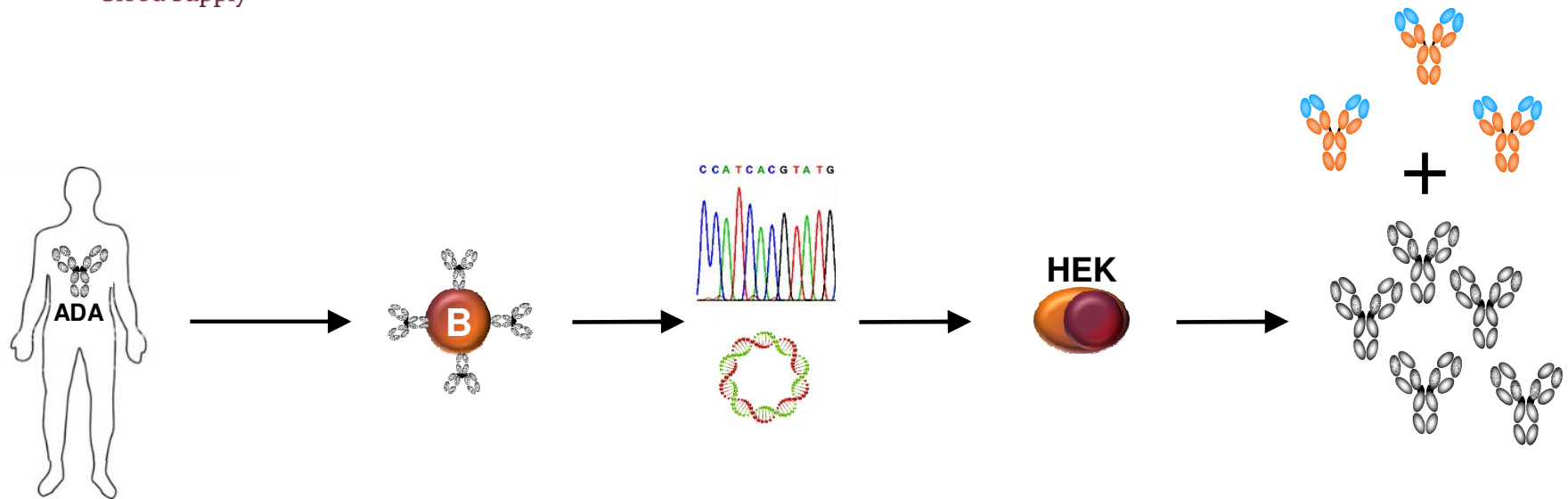


## Anti-drug antibodies to therapeutic antibodies are largely neutralizing



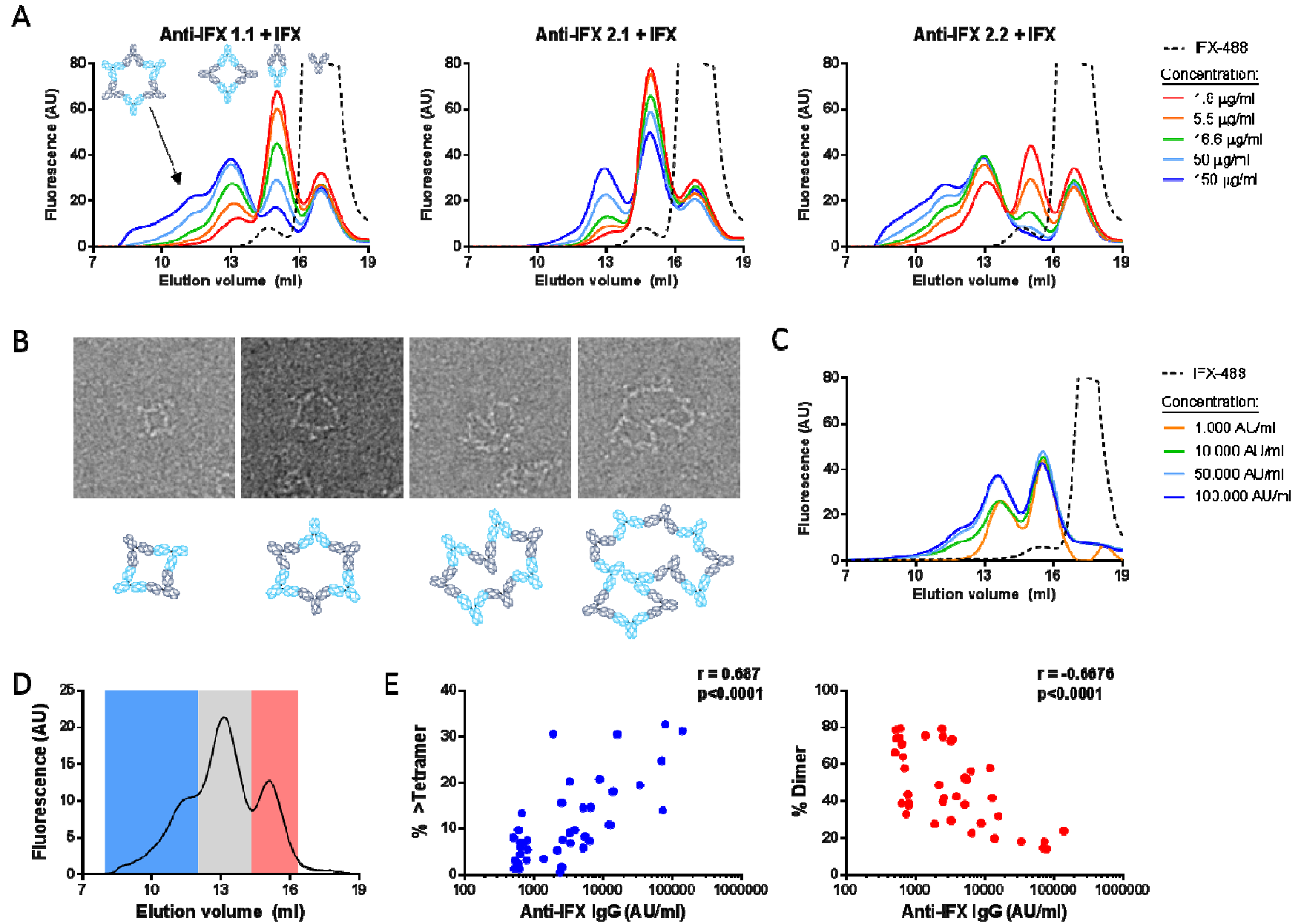
# Human monoclonal anti-infliximab antibodies

## Tools to investigate immune complexes

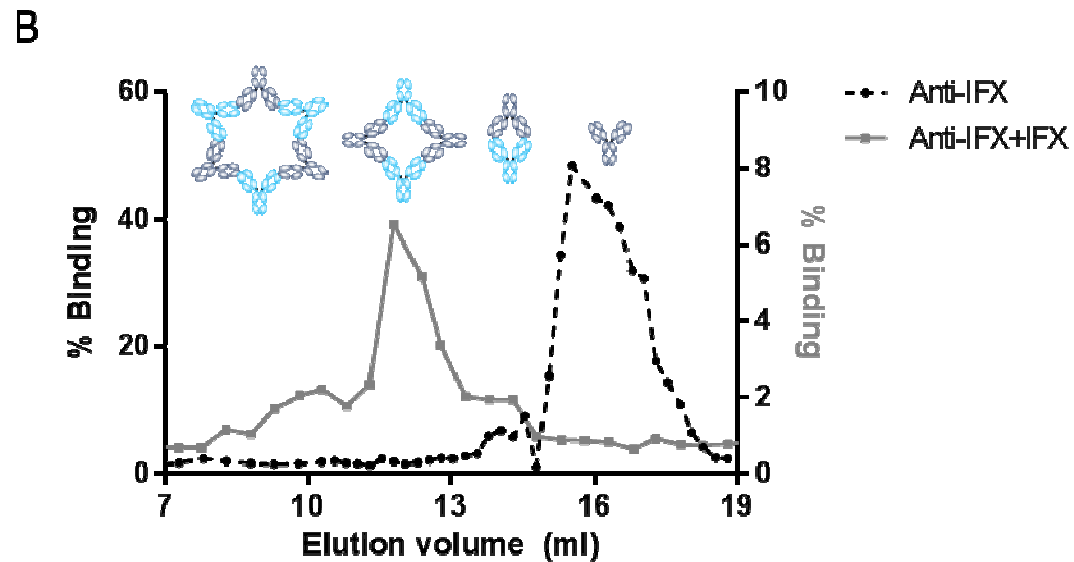
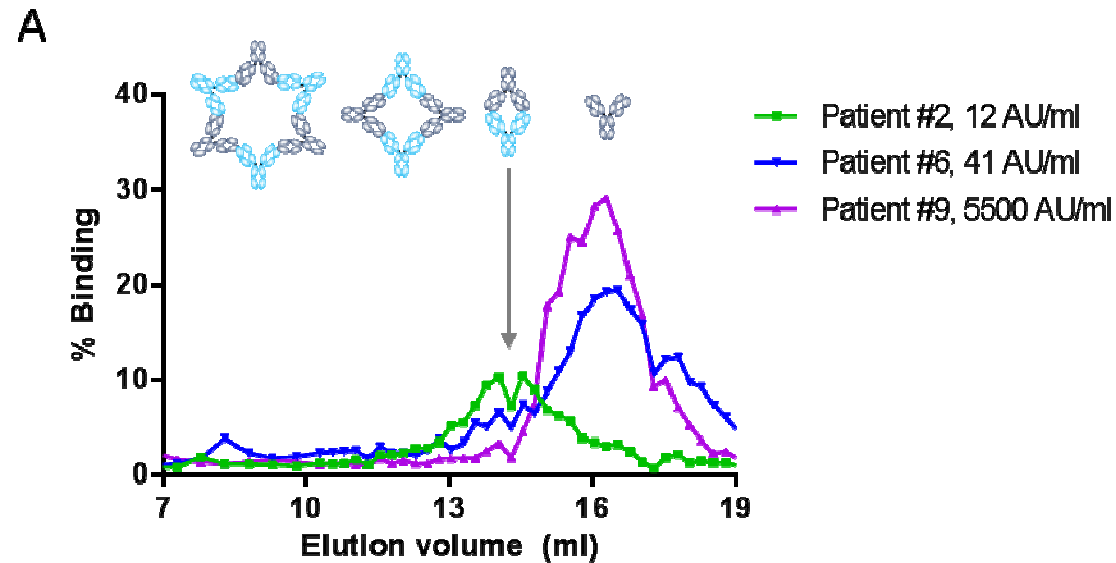


Clone	Neutralizing	Affinity	
		$K_D$ (pM)	SEM
1.1	Yes	76	4
1.2	Yes	280	85
1.3	Yes	1220	165
1.4	Yes	259	20
2.1	Yes	1670	171
2.2	Yes	134	4.5
2.3	Yes	460	35
2.4	Yes	143	10

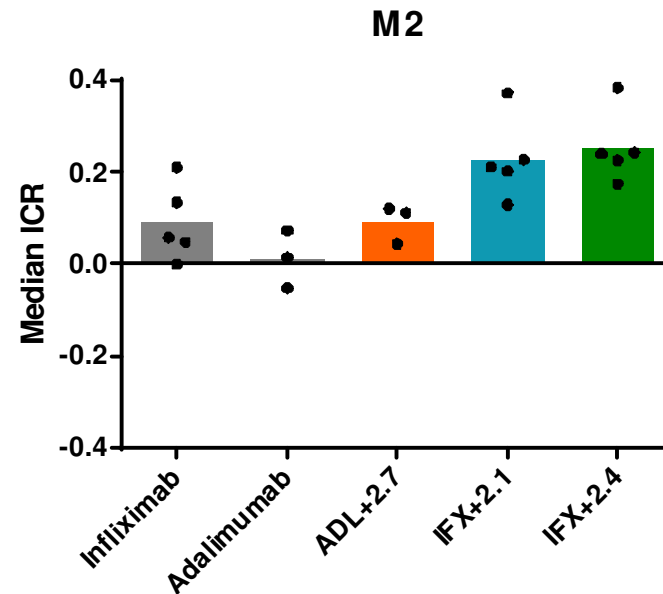
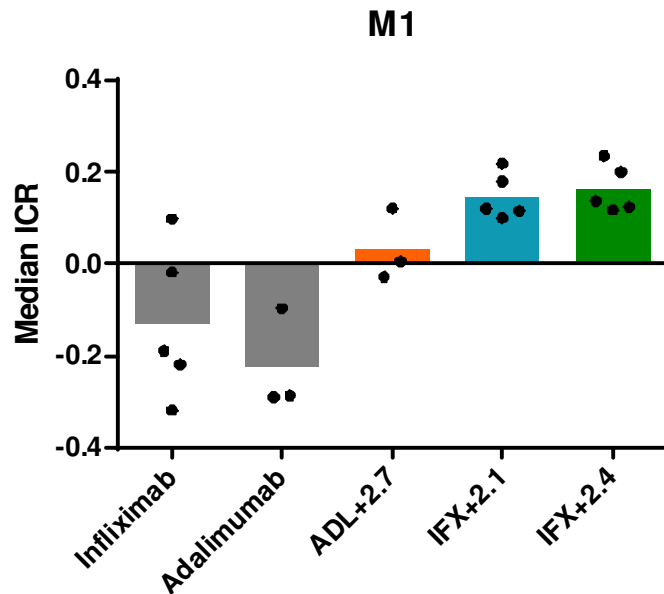
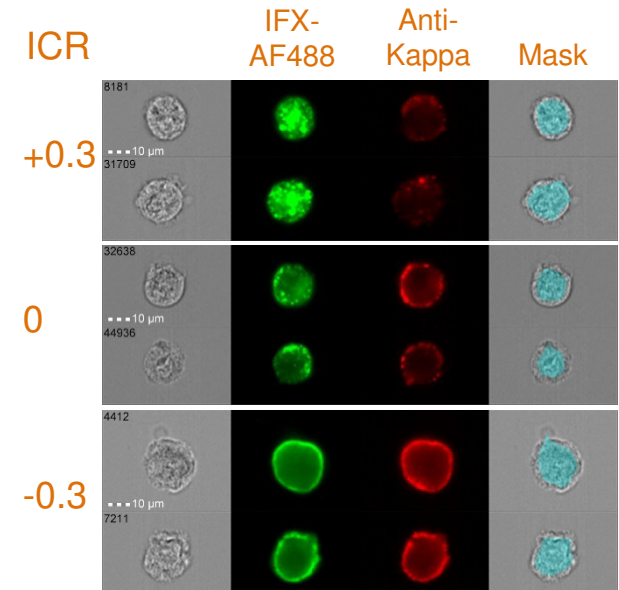
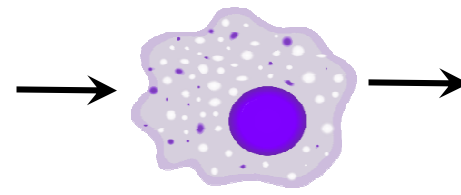
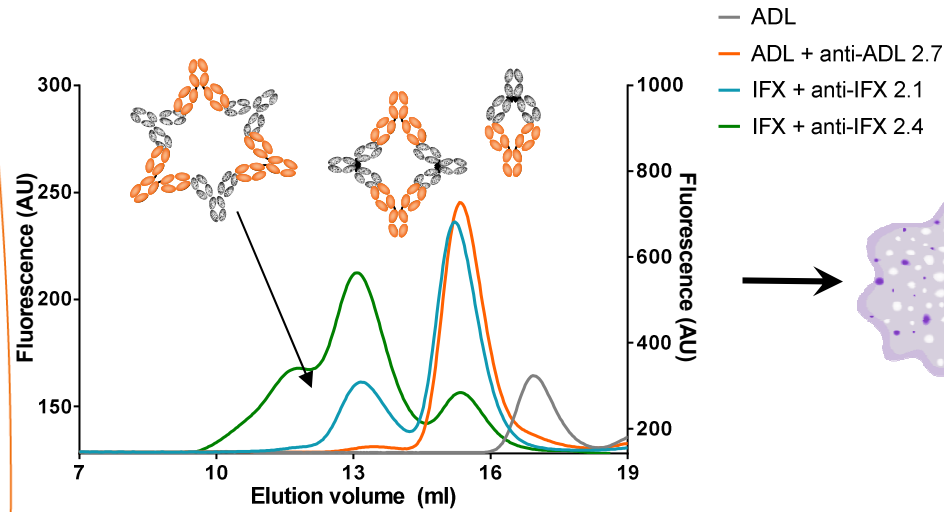
# Immune complex size is highly dependent on concentration



## Only small complexes and monomeric anti-infliximab antibodies are detected in patients at trough



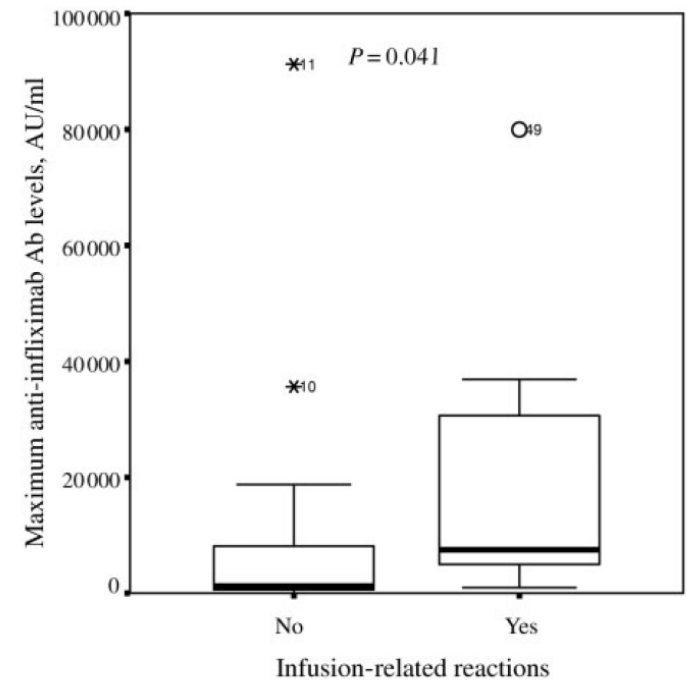
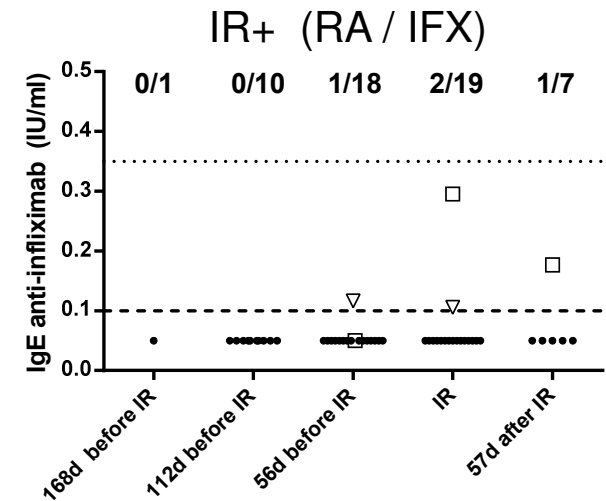
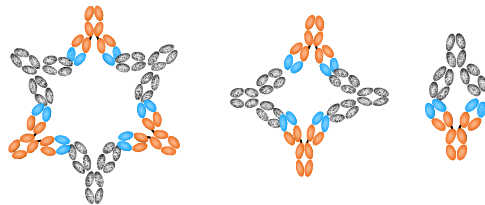
# Complexes larger than dimers are phagocytosed by macrophages



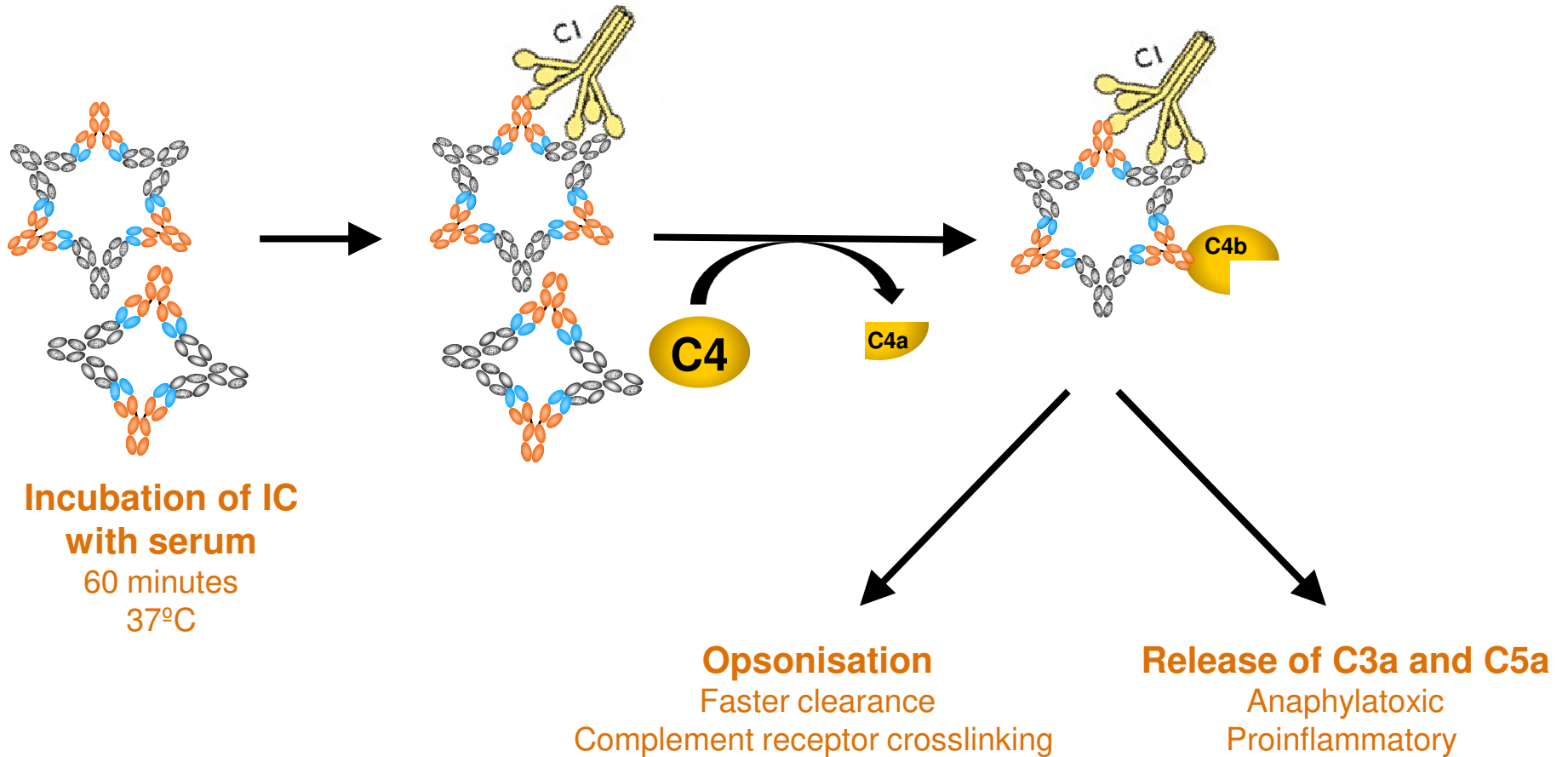
## Anti-idiotypic immune complexes & adverse events

## Adverse events

- ~7% of patients experience an infusion reaction (of varying severity) upon infliximab infusion
- Despite suggestions in literature, no clear association with anti-IFX IgE in patient sera
- anti-IFX IgE largely absent or present at levels <0.35 kIE/L. (*van Schie et al. Ann. Rheum. Dis. 2017*)
- High (IgG) ADA titer increases chance of infusion reactions (*Pascual-Salcedo et al., Rheumatology, 2011*)
- But also IgG-IFX immune-complex-mediated reactions uncommon
- Restricted Ab repertoire → small, ring-shaped complexes with limited immune activating capacity (*van Schie et al. Ann. Rheum. Dis. 2018*).

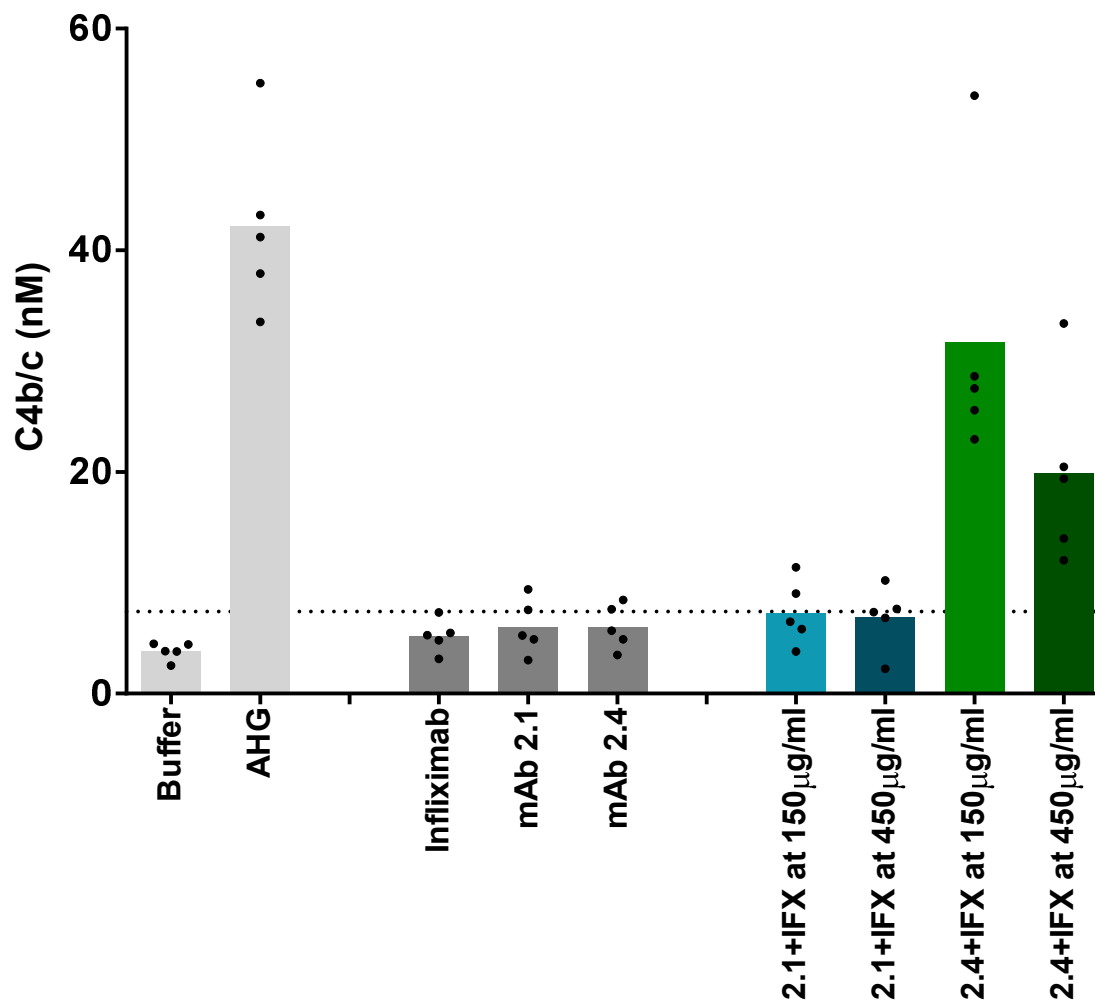
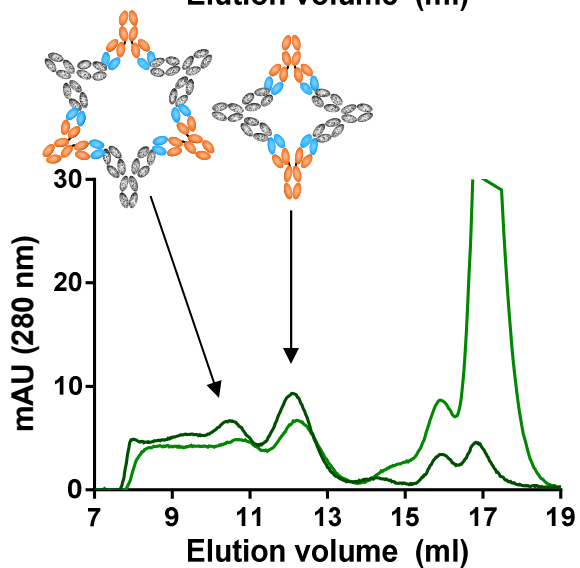
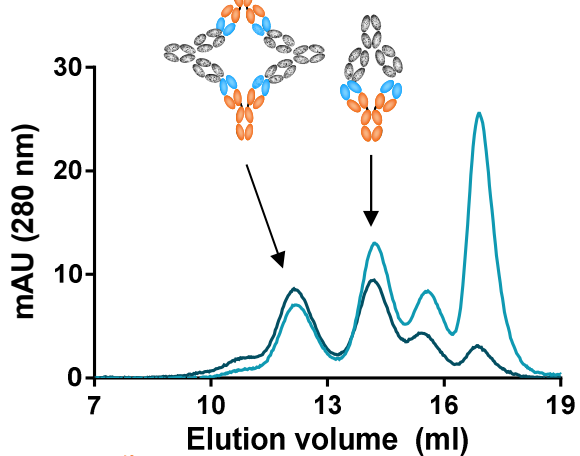


# Do immune complexes activate the complement system?

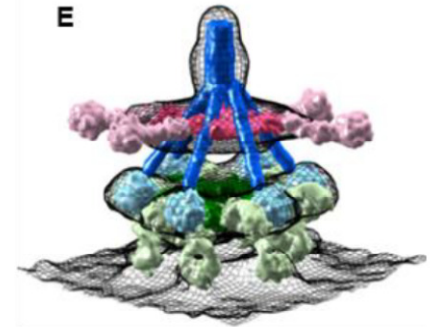
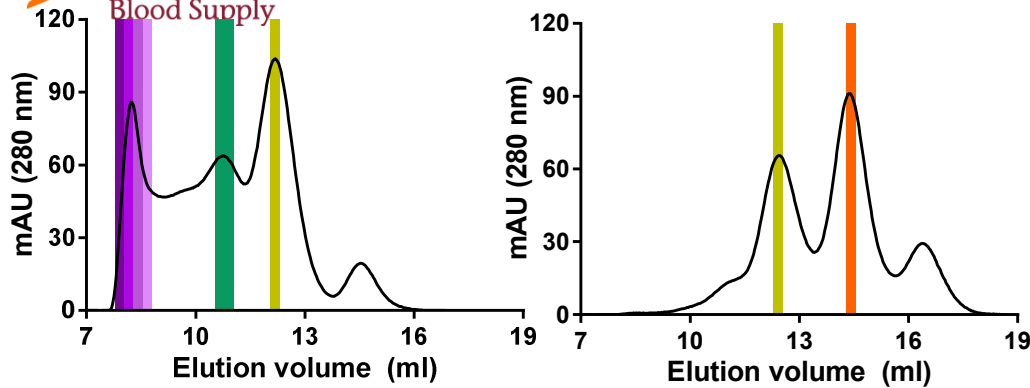




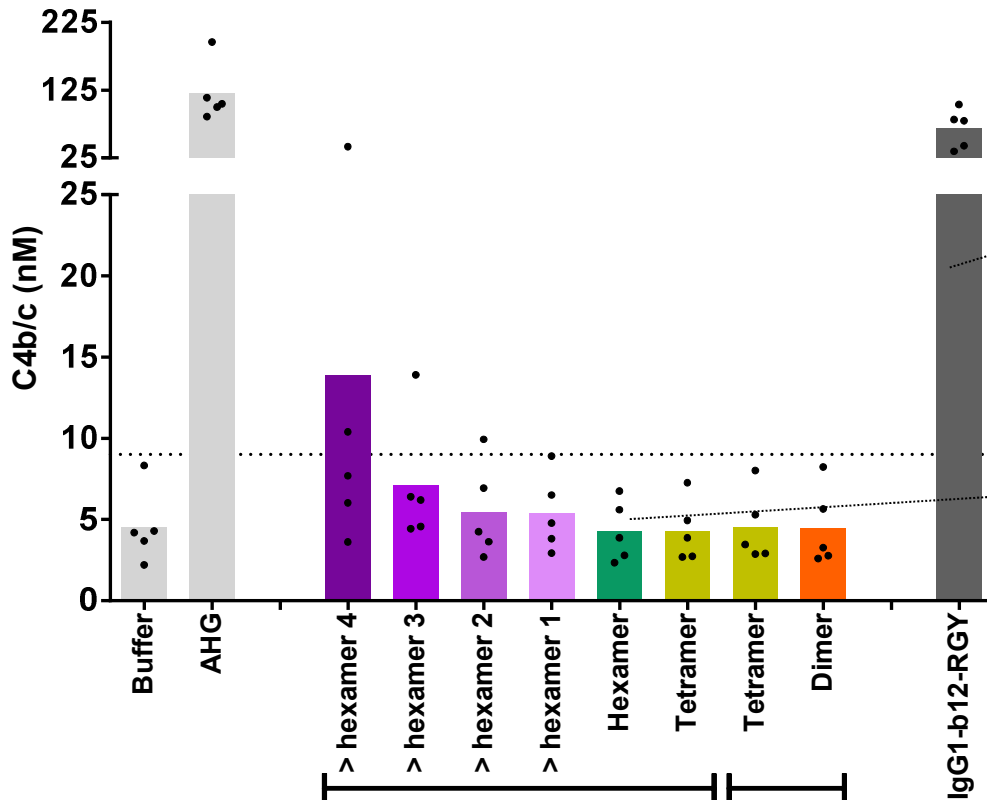
# Large anti-idiotypic immune complexes activate the complement system



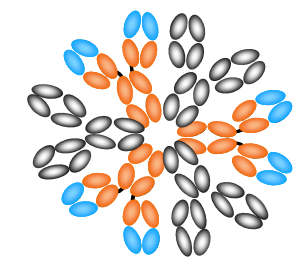
# Complexes larger than hexamers may activate complement



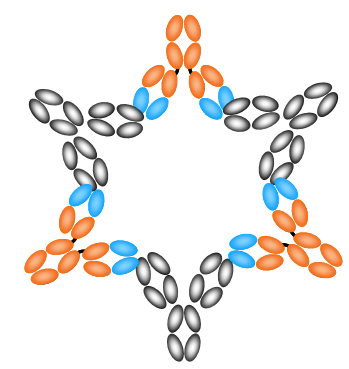
Diebolder et al., Science, 2014



mAb 2.4      mAb 2.1



IgG1-b12-RGY hexamer



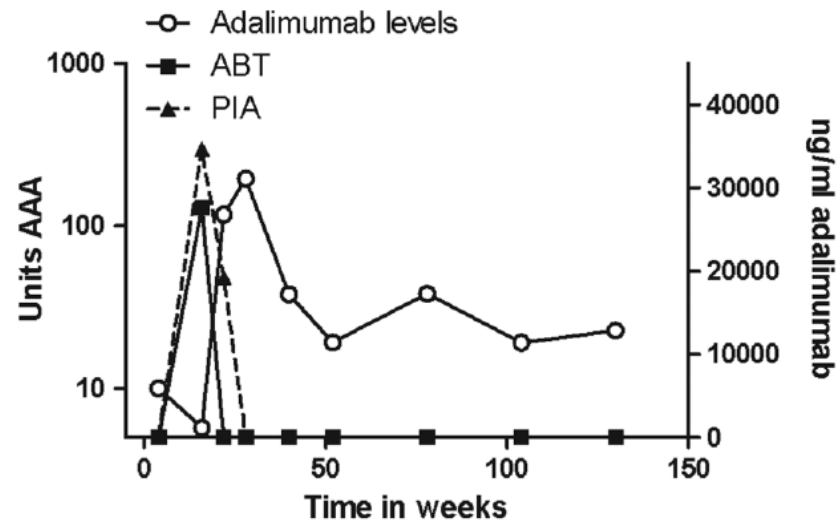
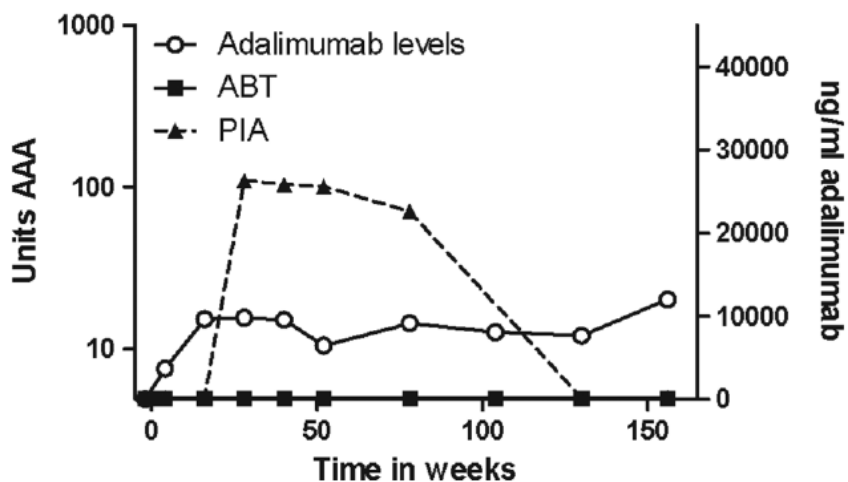
Anti-idiotypic hexamer

Prediction:

Dynamics of ADA response / transient responses

# Tolerance

More than a third of RA patients treated with adalimumab have a detectable, but transient antibody response



# IgM responses to adalimumab



# IgM responses to adalimumab

Target: TNF

(prediction of discontinuation?)

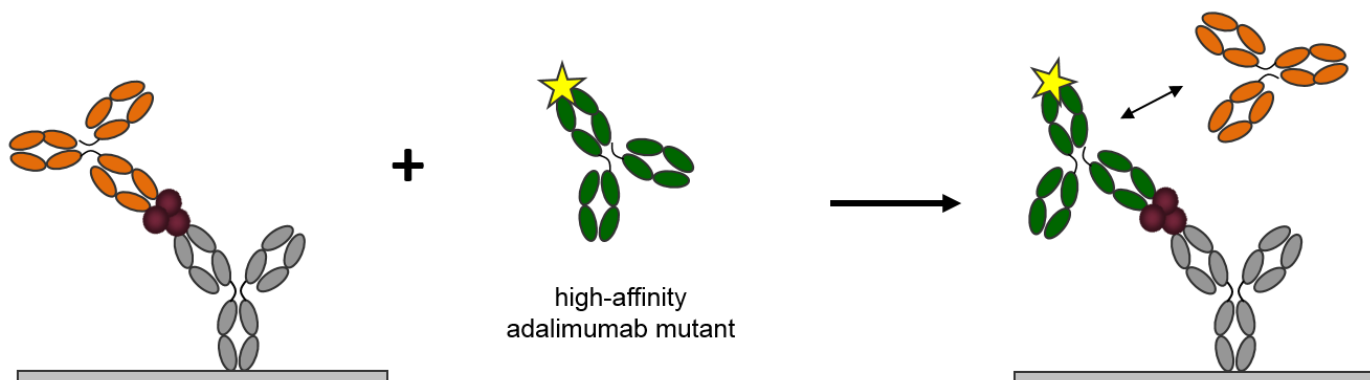
# Measurement of TNF in presence of TNFi

**A**

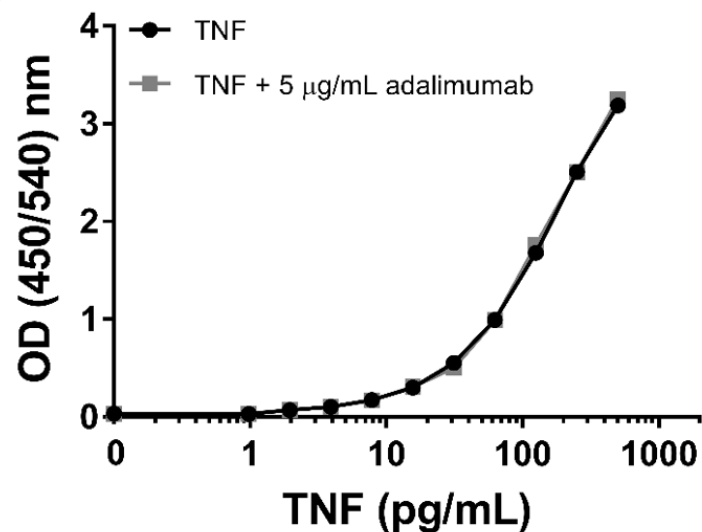
adalimumab

TNF

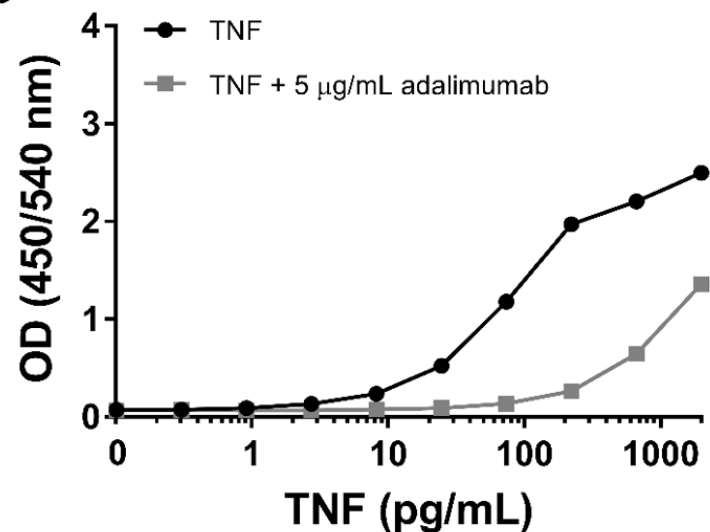
anti-TNF-7



**B**

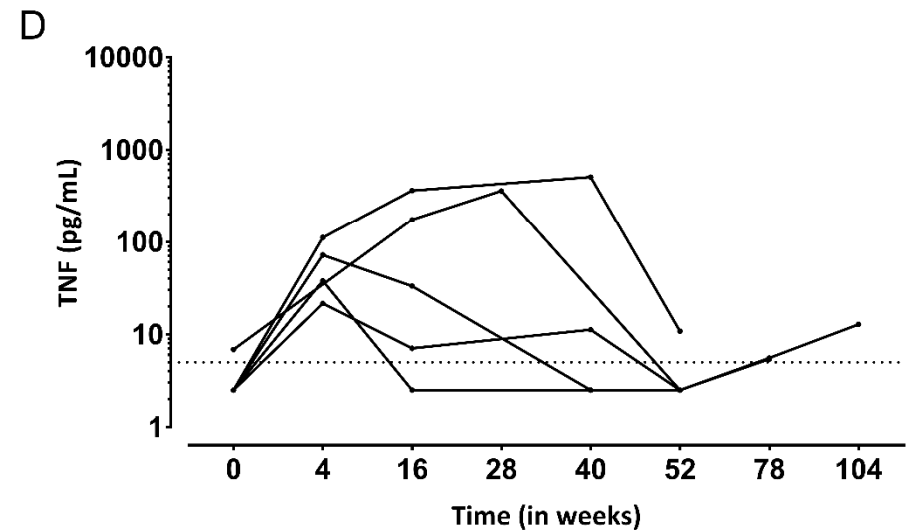
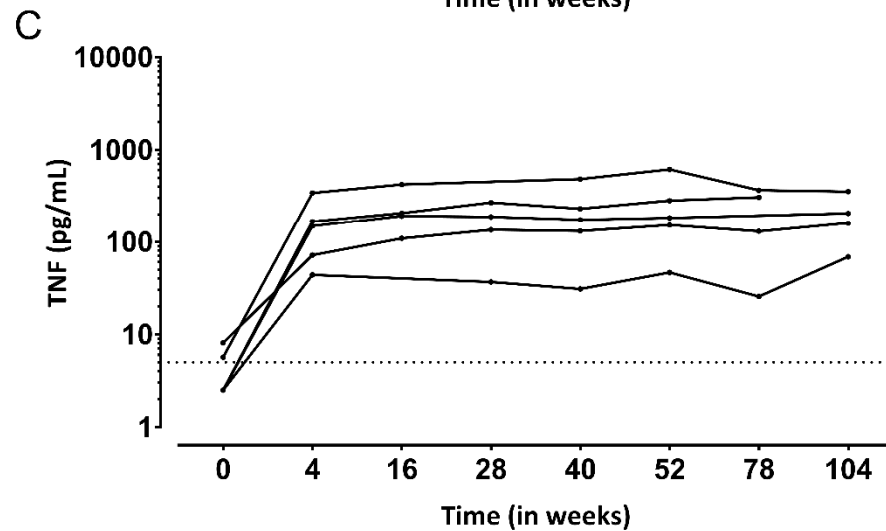
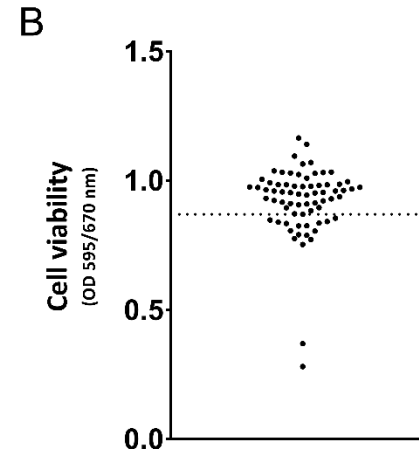
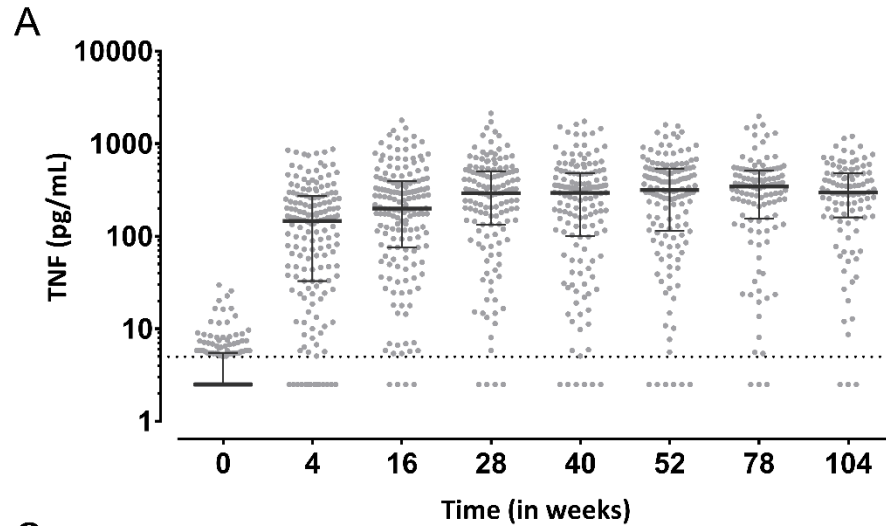


**C**



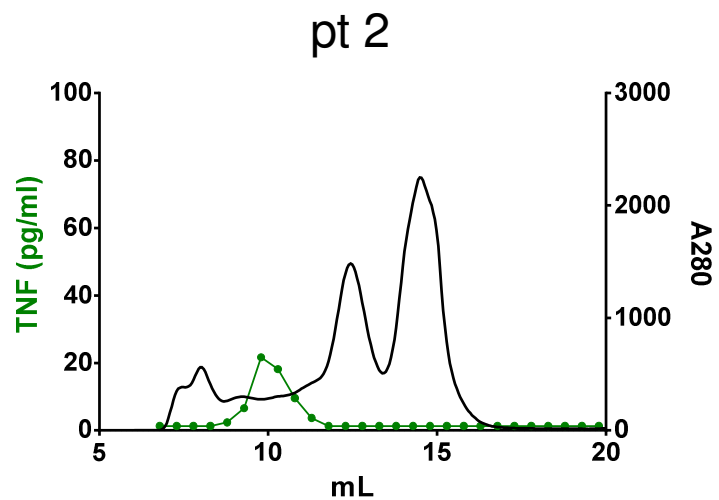
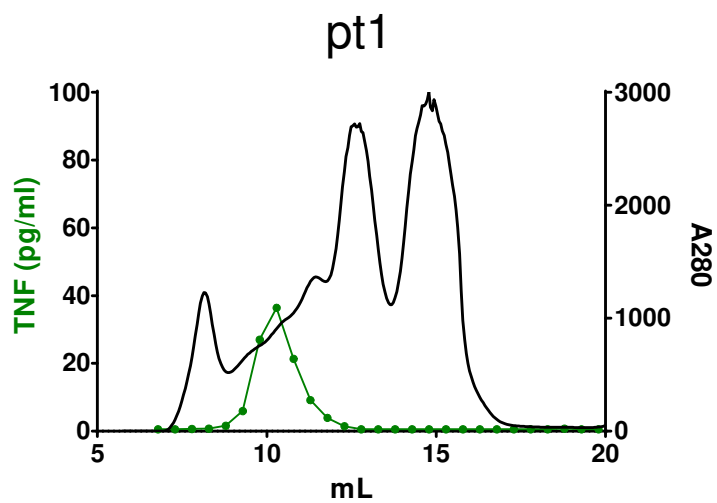
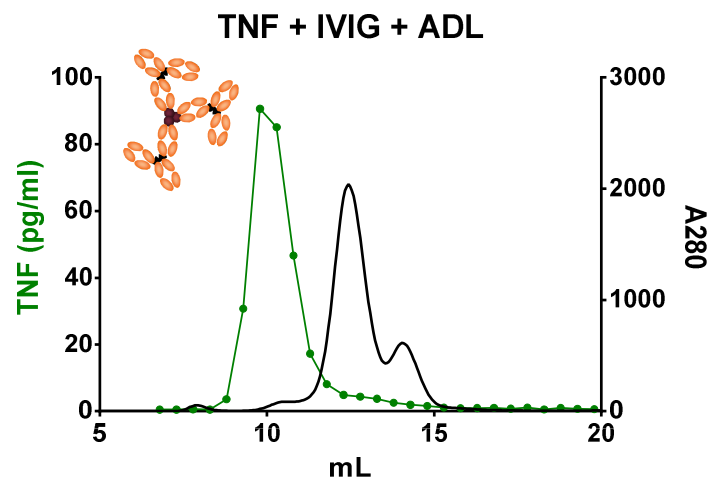
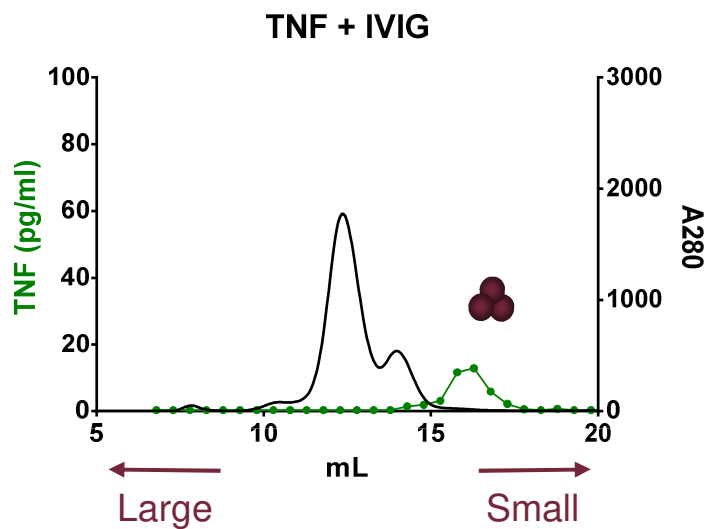


# TNF in adalimumab-treated RA patients

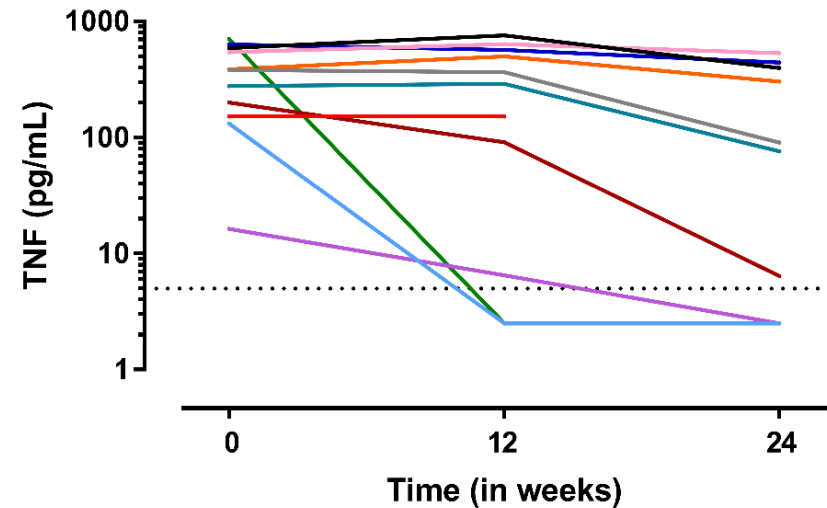
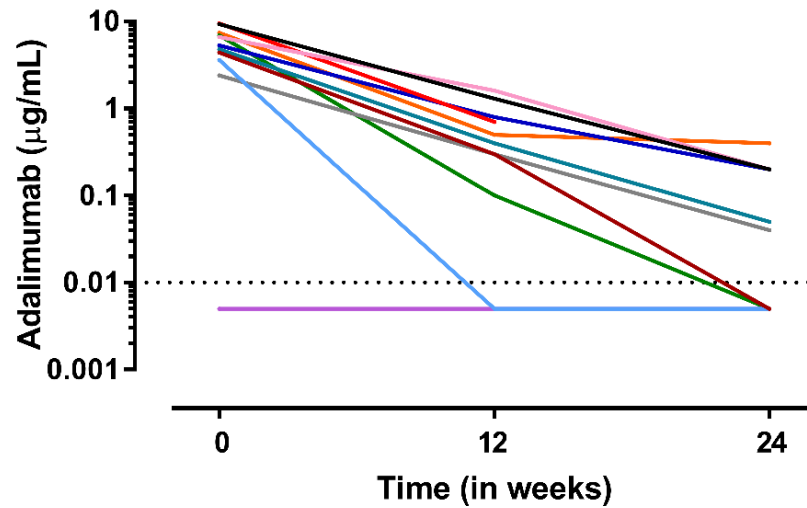


# TNF-adalimumab complexes

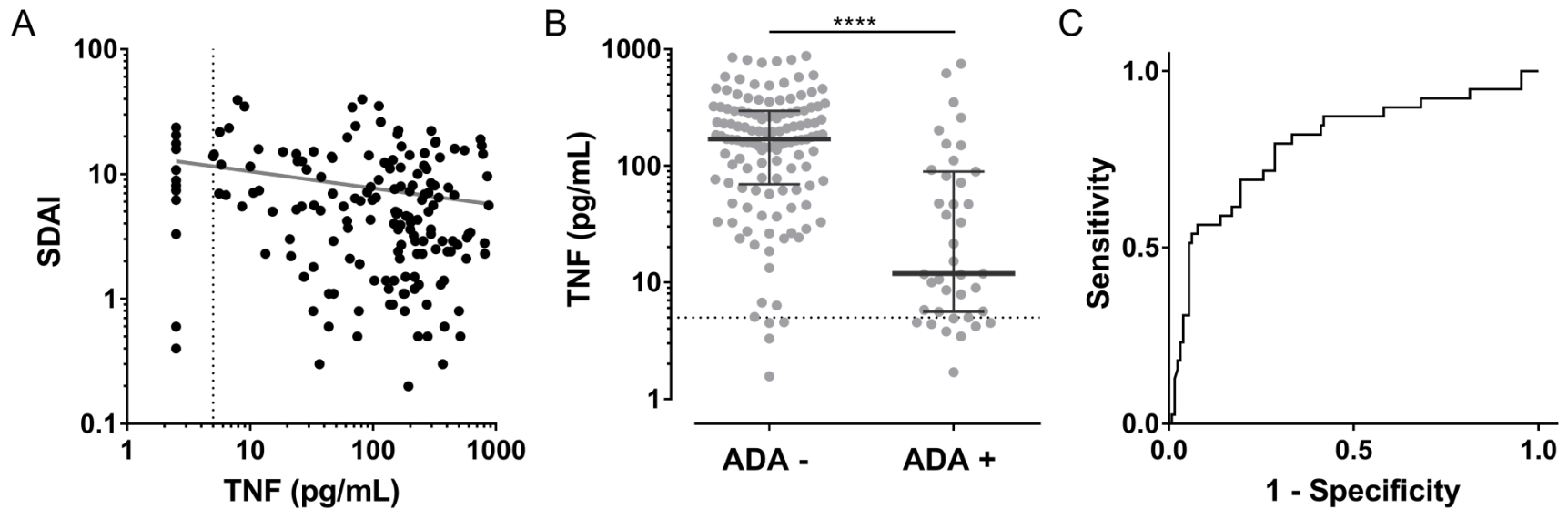
HPLC → Competition ELISA



# TNF and adalimumab levels upon discontinuation

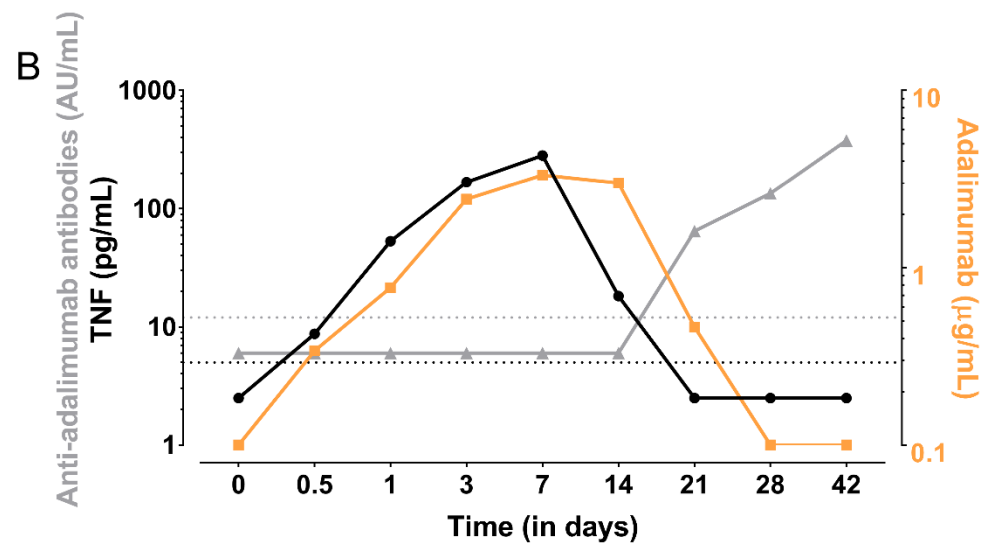
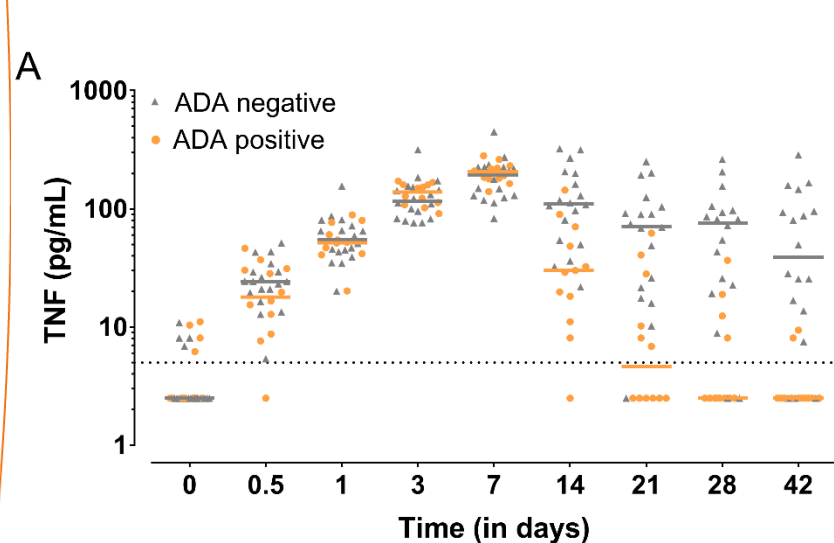


# Week 4 TNF levels associate with subsequent ADA formation



# Association TNF with antibody formation to adalimumab

Healthy volunteers – single dose adalimumab 40 mg (no MTX)





## Take home

- Impact of ADA on efficacy: related to PK
  - Measurement of ADA useful to support interpretation of PK
  - Both clearance & neutralization
  - Relation PK – efficacy: complex (not discussed here)
- Impact of ADA on adverse events: rare
  - Anti-idiotypic complexes → restricted immune activation
- Prediction of (clinically relevant) ADA: challenging
  - Transient responses: ADA measurements not always predictive
  - Markers to predict 'persistent' responses (TNF for adalimumab?)?



## Sanquin Acknowledgements

### **Sanquin Research**

Karin van Schie  
Pauline van Schouwenburg  
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