Biologics & risk in phase I studies

How do we do it better?

Dr Richard FitzGerald

Director, Clinical Research Unit, RLUH

Consultant Physician, CPT & GIM, RLUH

Molecular & Clinical Pharmacology, UoL

Richard.Fitzgerald@liverpool.ac.uk

UNIVERSITY OF LIVERPOOL

THE WOLFSON CENTRE FOR PERSONALISED MEDICINE

MRC Centre for Drug Safety Science

News Front Page
World
UK
England
Northern Ireland
Scotland
Wales
Business
Politics
Health
Education
Science & Environment
Technology
Entertainment
Also in the news
Video and Audio
Have Your Say
Magazine
In Pictures
Country Profiles
Special Reports

RELATED BBC SITES
SPORT
WEATHER
CBBC NEWSROUND
ON THIS DAY
EDITORS' BLOG

Last Updated: Wednesday, 15 March 2006, 22:37 GMT

E-mail this to a friend Printable version

Two drug trial men critically ill

Two men remain critically ill and four others are in a serious condition after suffering a violent reaction while taking part in a clinical drugs trial.

The six are being treated at Northwick Park hospital

All are still in intensive care in Northwick Park Hospital, north-west London, after falling ill on Monday.

Myfanwy Marshall told BBC News her boyfriend's body was badly swollen and she had been told he could die.

One of the drugs companies involved in the trial said it has apologised to the families of the men.

But relatives are said to be unhappy with the information given from the firm behind the anti-inflammatory drug.

The families had been given "mixed messages" during two meetings with pharmaceutical company TeGenero AG, which manufactures the drug and Parexel, which ran the trial, it was claimed.

Lawyer Ann Alexander, representing one of the critically ill men, told the BBC the companies had been asked whether any of the animals used to test the drug had died.

BBC London
Sport, travel, weather, things to do, features and much more

SEE ALSO:
- 'They say he needs a miracle' 15 Mar 06 | Health
- Making a career out of drug tests 15 Mar 06 | London
- Q&A: Drug trials 15 Mar 06 | Health
- Six taken ill after drug trials 15 Mar 06 | London
- Confusion over drug trial rights 26 Feb 06 | Health
- Woman died on cannabis drug trial 12 Dec 05 | South Yorkshire

RELATED BBC LINKS:
- Clinical trials

RELATED INTERNET LINKS:
- Department of Health
- North West London Strategic Health Authority
- North West London Hospitals NHS Trust
- Parexel
- MHRA
- Medical Research Council

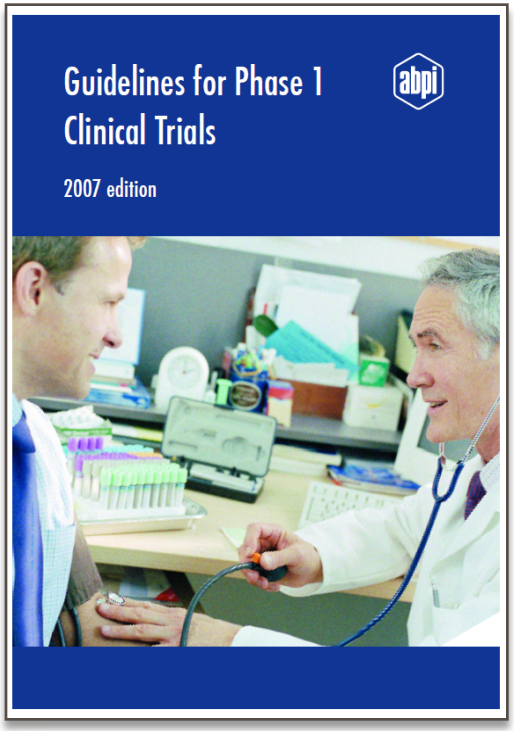
MHRA

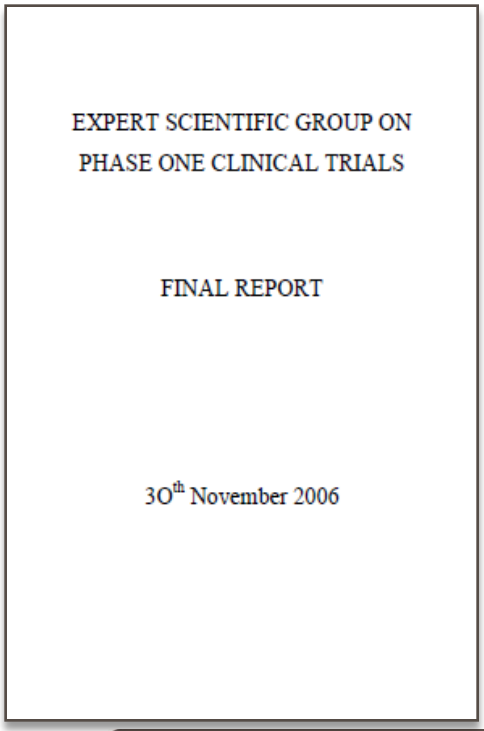

The NEW ENGLAND JOURNAL of MEDICINE

BRIEF REPORT

Cytokine Storm in a Phase 1 Trial of the Anti-CD28 Monoclonal Antibody TGN1412

Ganesh Suntharalingam, F.R.C.A., Meghan R. Perry, M.R.C.P., Stephen Ward, F.R.C.A., Stephen J. Brett, M.D., Andrew Castello-Cortes, F.R.C.A., Michael D. Brunner, F.R.C.A., and Nicki Panoskaltsis, M.D., Ph.D.




EXPERT SCIENTIFIC GROUP ON PHASE ONE CLINICAL TRIALS

FINAL REPORT

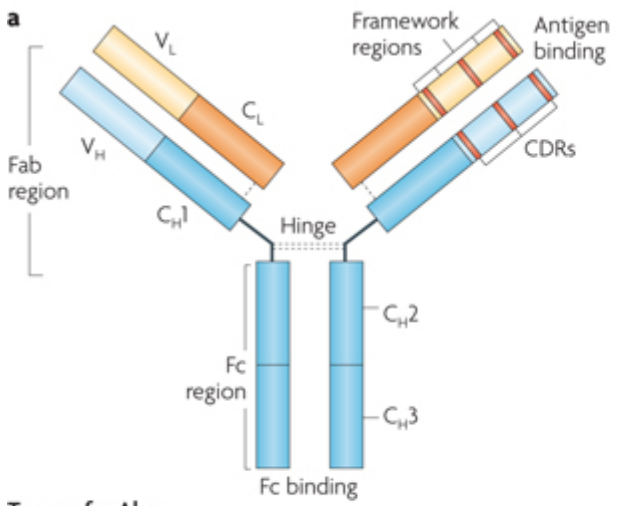
30th November 2006

UNIVERSITY OF LIVERPOOL

THE WOLFSON CENTRE FOR PERSONALISED MEDICINE

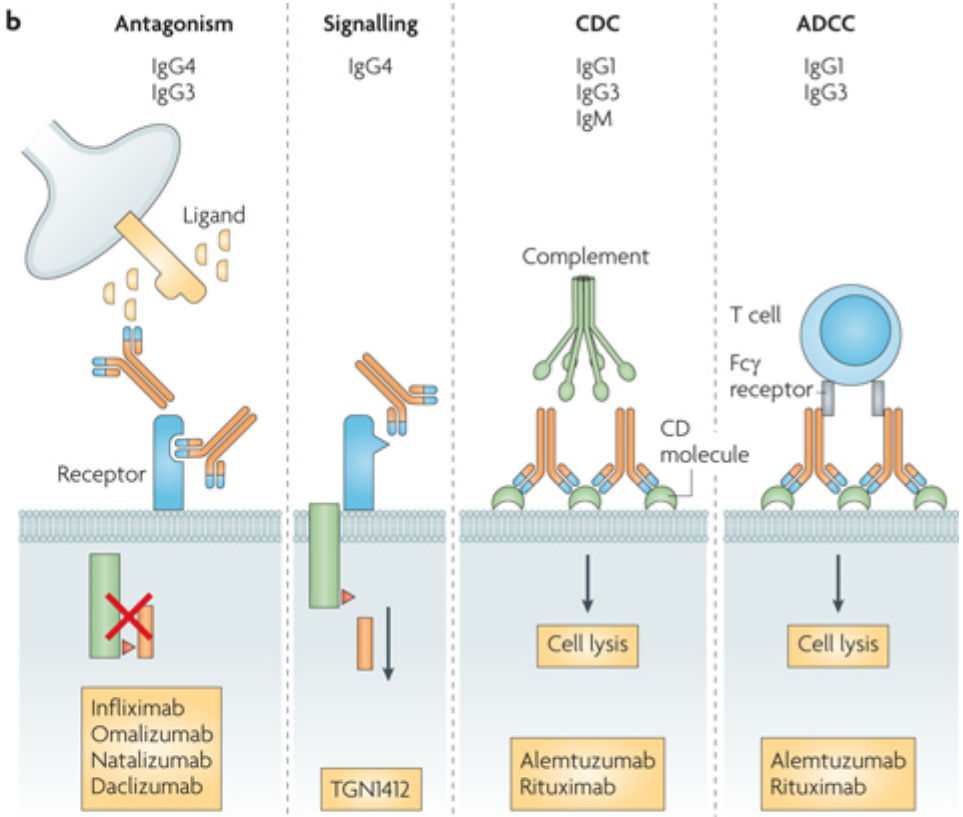
MRC Centre for Drug Safety Science

Mechanism of action



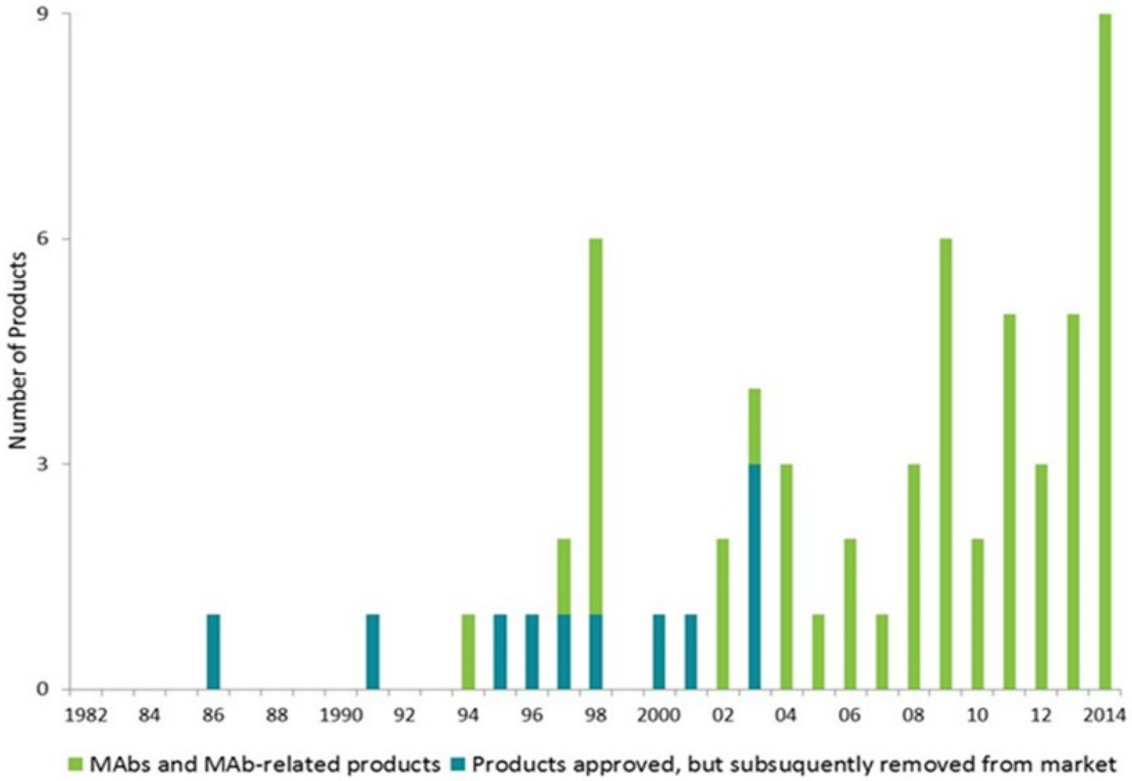
Types of mAbs

Murine	Entirely murine amino acids	'o' = mouse e.g. muromonab
Chimeric	Human constant (C) + murine variable (V) regions	'xi' = chimeric e.g. rituximab
Humanized	Murine complementarity determining regions (CDRs)	'zu' = humanized e.g. alemtuzumab
Human	Entirely human amino acids	'u' = human e.g. adalimumab



Nature Reviews | Drug Discovery

Bigger, better, more?



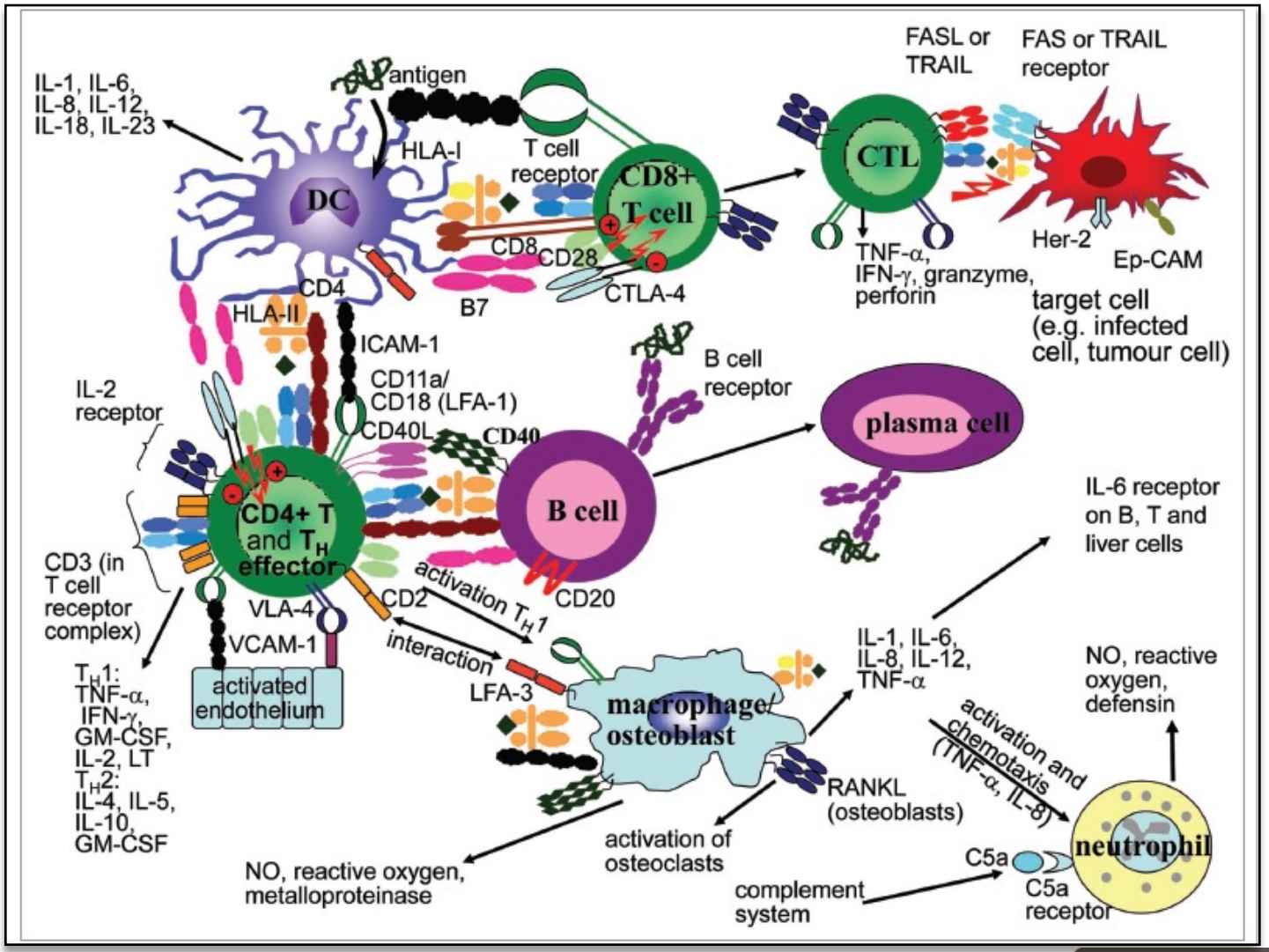
The graph displays the number of monoclonal antibody products first approved for commercial sale in the US or Europe each year since 1982. The totals include all monoclonal antibody and antibody-related products. Products approved but subsequently removed from the market are denoted in blue; products currently marketed are denoted in green. For 2014, the figure includes the total number of products approved as of December 31.

TABLE 1. mAbs and Fc-Fusion Proteins Approved by FDA and/or EMA for Inflammatory Disease Indications

INN name	Trade name	Company	Species/isotype	Target	Inflammatory indication(s)	MoA
Muromonab	**Orthoclone-OKT3®**	Janssen-Cilag	Mouse IgG2a	CD3	Organ rejection (renal, heart, liver)	Inhibits T cell function.
Daclizumab	**ǂZenapax®**	Roche	Humanized IgG1	IL-2R	Organ rejection (renal)	Inhibits T cell function
Rituximab	**Rituxan™, Mabthera™**	Biogen-IDEC, Roche	Chimeric IgG1	CD20	RA*	B cell depletion by ADCC, CDC and apoptosis
Basilixumab	**Simulect®**	Novartis	Chimeric IgG1	IL-2R	Organ rejection (renal)	Inhibits T cell function
Infliximab	**Remicade®**	Johnson & Johnson	Chimeric IgG1	TNFα	RA, CrD, UC, AS, Ps, PsA	Blocks TNFα -mediated leukocyte migration, inhibition of apoptosis, macrophage, osteoclast, and EC activation
Etanercept	**Enbrel®**	Amgen, Pfizer	Human TNFR-FcIgG1	TNFα	RA, Ps, CrD	Blocks TNFα -activity
Alemtuzumab	**Campath®**	Genzyme, Bayer	Humanized IgG1	CD52	RA, MS	Depletes leukocytes (T and B cells monocytes, macrophages) by ADCC
Adalimumab	**Humira™/ Trudexa**	AbbVie	Human IgG1	TNFα	RA, CrD, AS, Ps, PsA, JIA	Blocks TNFα -activity
Omalizumab	**Xolair®**	Genentech, Roche, Novartis	Humanized IgG1	IgE	Allergic asthma	Blocks IgE binding to, and cross-linking of, FcεRI on mast cells
Efalizumab	**ǂRaptiva®**	Genentech, Merck-Serono	Humanized IgG1	CD11a (LFA-1)	Ps	Blocks inflammatory T cell migration
Alefacept	**Amevive®**	Astellas	Human LFA-3-FcIgG1	CD2	Ps	Inhibition of T cell function (binds CD2 on T cells blocking interaction with LFA-3 on APC).
Natalizumab	**Tysabri®**	Biogen IDEC, Elan	Humanized IgG4	CD49d (VLA-4)	MS, CrD	Blocks inflammatory T cell migration
Abatacept	**Orencia®**	Bristol-Myers Squibb	Human CTLA4-FcIgG1	CD80, CD86	RA, JIA	Inhibition of T cell function (binds to CD80 & CD86 on APCs leading to blocking of CD28 interaction and T cell activation)
Eculizumab	**Soliris®**	Alexion	Humanized IgG2/4	C5	PNH	Blocks complement C5, inhibiting its cleavage to C5a and C5b and generation of membrane attack complex C5b-C9
Certolizumab pegol	**Cimzia®**	UCB	Humanized Fab-PEG	TNFα	CrD	Blocks TNFα activity
Golimumab	**Simponi™**	Johnson & Johnson	Human IgG1	TNFα	RA, AS, PsA	Blocks TNFα activity
Canakinumab	**Ilaris®**	Novartis	Human IgG1	IL-1β	CAPS	Blocks IL-1β-mediated leukocyte migration, macrophage & osteoclast activation, DC activation, Th17 differentiation
Tociluzumab	**Actemra®, RoActemra®**	Roche, Chugai	Humanized IgG1	IL-6R	RA	Blocks IL-6-mediated B/T cell and osteoclast activation, inhibition of T cell apoptosis, Th17 differentiation
Ustekinomab	**Stelara™**	Johnson & Johnson	Human IgG1	IL-12/23 (p19)	Ps	Blocks IL-12-mediated TH1/NK cell activation & TNFα/IFN-γ release; blocks IL-23-mediated TH17 cell expansion and IL-17/IL-22 release
Belimumab	**Benlysta Lymphostat B**	Human Genome Sciences/Glaxo Smith Kline	Human IgG1	BLyS (BAFF)	SLE	B cell depletion by apoptosis (Block Blys binding to BCMA, BAFFR and TACI required for B cell maturation/survivial)
Belatacept	**Nulojix®**	Bristol-Myers Squibb	Human CTLA-4-IgGFc-IgG4	CD80, CD86	Organ rejection (renal)	Inhibition of T cell function (binds to CD80 & CD86 on APCs leading to blocking of CD28 interactions & T cell activation)

TABLE 2. mAbs in Phase III Clinical Trials for Inflammatory Disease Indications

INN name	Company	Species	Target	Indication(s)	MoA
Gevokizumab	Xoma-Servier	Humanized IgG2	IL-1β	Uveitis (non-infectious)	Blocks IL-1β activity
Mepolizumab	Glaxo Smithkline	Humanized IgG1	IL-5	Asthma (eosinophilic)	Inhibits IL-5-mediated eosinophil maturation, activation and migration (binds IL-5 thereby blocking binding to IL-5R on eosinophils).
Reslizumab	Teva (Cephalon/ Ception Therapeutics)	Humanized IgG4	IL-5	Asthma (eosinophilic)	Inhibits IL-5 activity
Benralizumab	Astra Zeneca-Medimmune	Humanized IgG1 (afucosylated)	IL-5Rα	Asthma (eosinophilic)	Inhibits IL-5 activity
Sarilumab	Sanofi (from Regeneron)	Human IgG1	IL-6Rα	RA	Inhibits IL-6 activity
Sirukumab	Janssen	Human IgG1	IL-6	RA	Inhibits IL-6 activity
Lebrikizumab	Roche	Humanized IgG4	IL-13	asthma	Blocks IL-13-mediated B cell proliferation/IgE production, mast cell and eosinophil recruitment, M2 macrophage activation, eotaxin production, collagen synthesis, mucus production and bronchoconstriction
Secukinumab	Novartis	Human IgG1	IL-17A	Ps, RA/PsA, AS	Blocks IL-17A-mediated pro-inflammatory cytokine/chemokine production, granulocyte and monocyte mobilization, TNFα/IL-1β production, ICAM-1 expression, DC maturation, EC and osteoclast activation
Ixekizumab	Eli Lilly	Humanized IgG4	IL-17A	Ps, PsA, RA	Blocks IL-17A activity
Brodalumab	Amgen	Human IgG2	IL-17RA	Ps	Blocks IL-17A, E,F-activity
Ocrelizumab	Roche	Humanized IgG1	CD20	MS	Induce B cell depletion by ADCC, CDC and apoptosis induction
Epratuzumab	UCB	Humanized IgG1	CD22	SLE	Induces downregulation of the B cell receptor, inhibition of B cell activation and B cell reduction
Atacicept	Merck-Serono	Human TACI ECD-FcIgG1	BlyS (BAFF)	SLE	Induces B cell depletion by apoptosis induction resulting in inhibition of B cell function (blocks Blys and APRIL required for B cell maturation & survivial)
Tabalumab	Eli Lilly	Human IgG4	BLyS (BAFF)	SLE	Induces B cell depletion by promotion of apoptosis (blocks Blys binding to BCMA, BAFFR and TACI required for B cell maturation and survivial)
Vedolizumab*	Takeda- Millenium	Humanized IgG1	α4β7	UC, CrD	Inhibits lymphocyte migration to gut
Itolizumab	Biocon	Humanized IgG1 kappa	CD6	Ps; RA, MS	Inhibits CD6-mediated T cell activation, cytokine production and migration


IL-1, IL-6, IL-8, IL-12, IL-18, IL-23
antigen
HLA-I
T cell receptor
DC
CD8+ T cell
CD8
CD28
CTLA-4
B7
CD4
HLA-II
ICAM-1
CD11a/ CD18 (LFA-1)
CD40L
CD40
IL-2 receptor
CD4+ T and T_H effector
CD3 (in T cell receptor complex)
T_H1: TNF-α, IFN-γ, GM-CSF, IL-2, LT
T_H2: IL-4, IL-5, IL-10, GM-CSF
VLA-4
VCAM-1
activated endothelium
CD2
activation T_H1
interaction
LFA-3
B cell
B cell receptor
CD20
plasma cell
FASL or TRAIL
FAS or TRAIL receptor
CTL
TNF-α, IFN-γ, granzyme, perforin
Her-2
Ep-CAM
target cell (e.g. infected cell, tumour cell)
IL-6 receptor on B, T and liver cells
IL-1, IL-6, IL-8, IL-12, TNF-α
NO, reactive oxygen, defensin
macrophage/ osteoblast
activation and chemotaxis (TNF-α, IL-8)
RANKL (osteoblasts)
activation of osteoclasts
NO, reactive oxygen, metalloproteinase
neutrophil
C5a
C5a receptor
complement system

A new paradigm

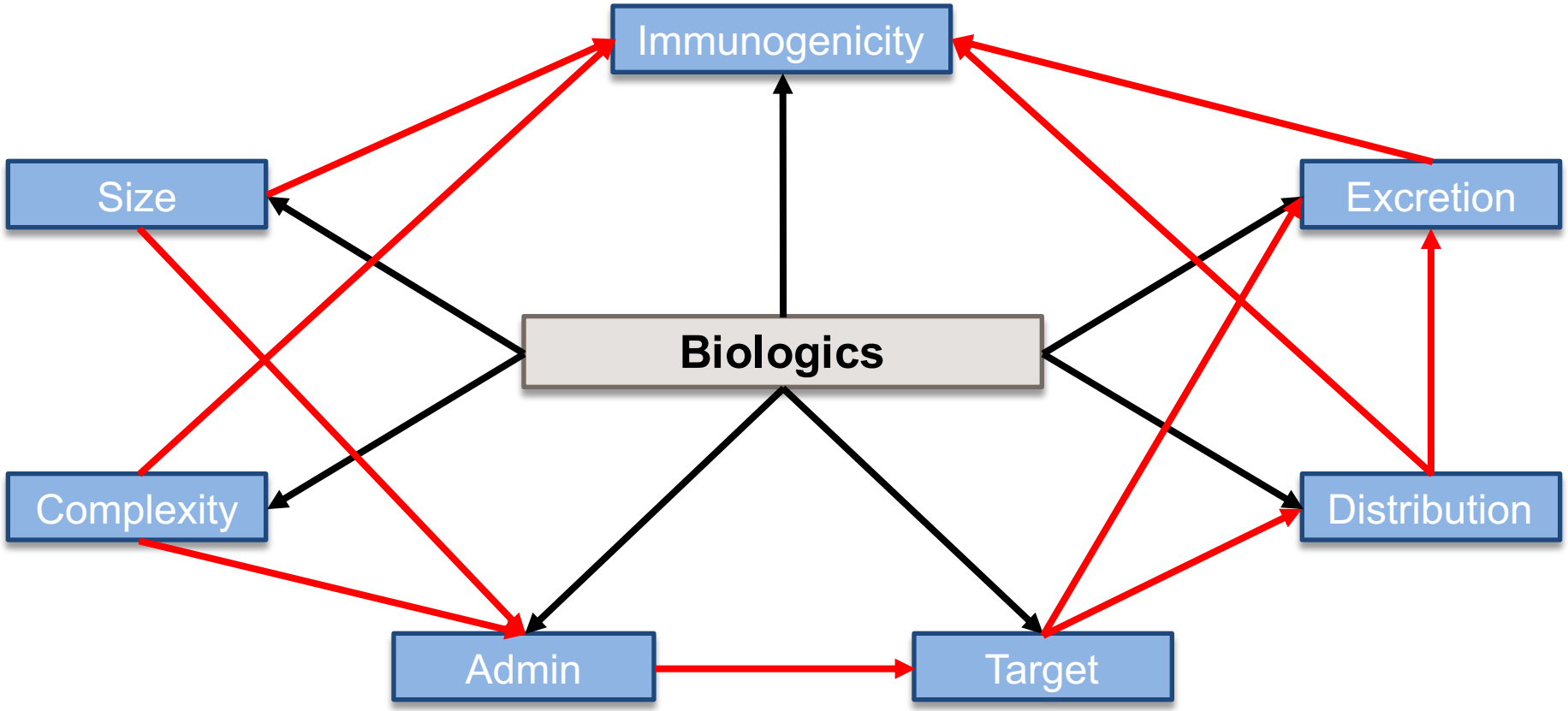
- Biologics are not small molecules

Biologics – Safer? Or not?

- Very target specific
- Less potential for off-target effects
- 'Simpler' metabolic pathway (generally)

Risk of exaggeration of on-target effect?
Characterisation of biological pathway?
Comparison between animal species and human?

Small molecule vs biologics



Size & Complexity - risks

- Very large molecules
  - 1 kDa to 1000 kDa
- Complex biochemistry
  - Protein folding
  - PEGylation
  - Disulphide bridges
  - Phosphorylation, sulphation, acetylation, carboxylation

Unpredictable in vivo behaviour
Risk of interaction with unexpected targets
Immunogenicty

Pharmacokinetics - risk

- Slow absorption (tmax = days if SC dose)
- Apparent small distribution
  - 2-4L typical
  - Beware paracellular or transcellular transport (endocytosis)
- Excretion via protein degradation
  - Lysosymal
  - Non-linear PK is typical
  - No effect from renal / hepatic function
  - Beware smaller proteins

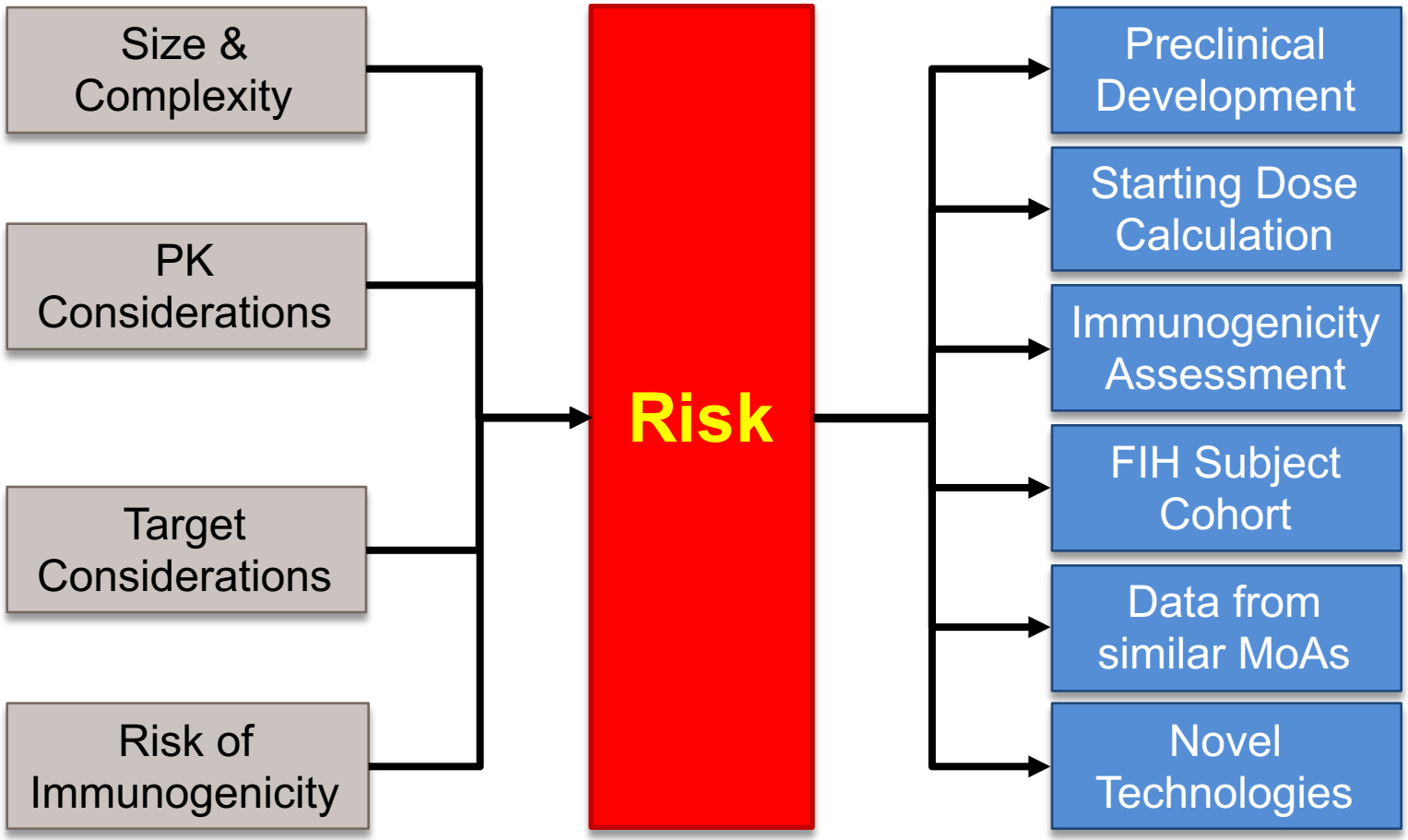
Target - risk

- Characterisation of target
  - Distribution
  - Effect + downstream consequence
- Cell based target?
  - Affects kinetics of IMP (higher apparent clearance at lower doses)
- Soluble target?
  - Affects kinetics of IMP (saturation kinetics)
- Target expression in disease?
  - Choice of population?

Immunogenicity - risk

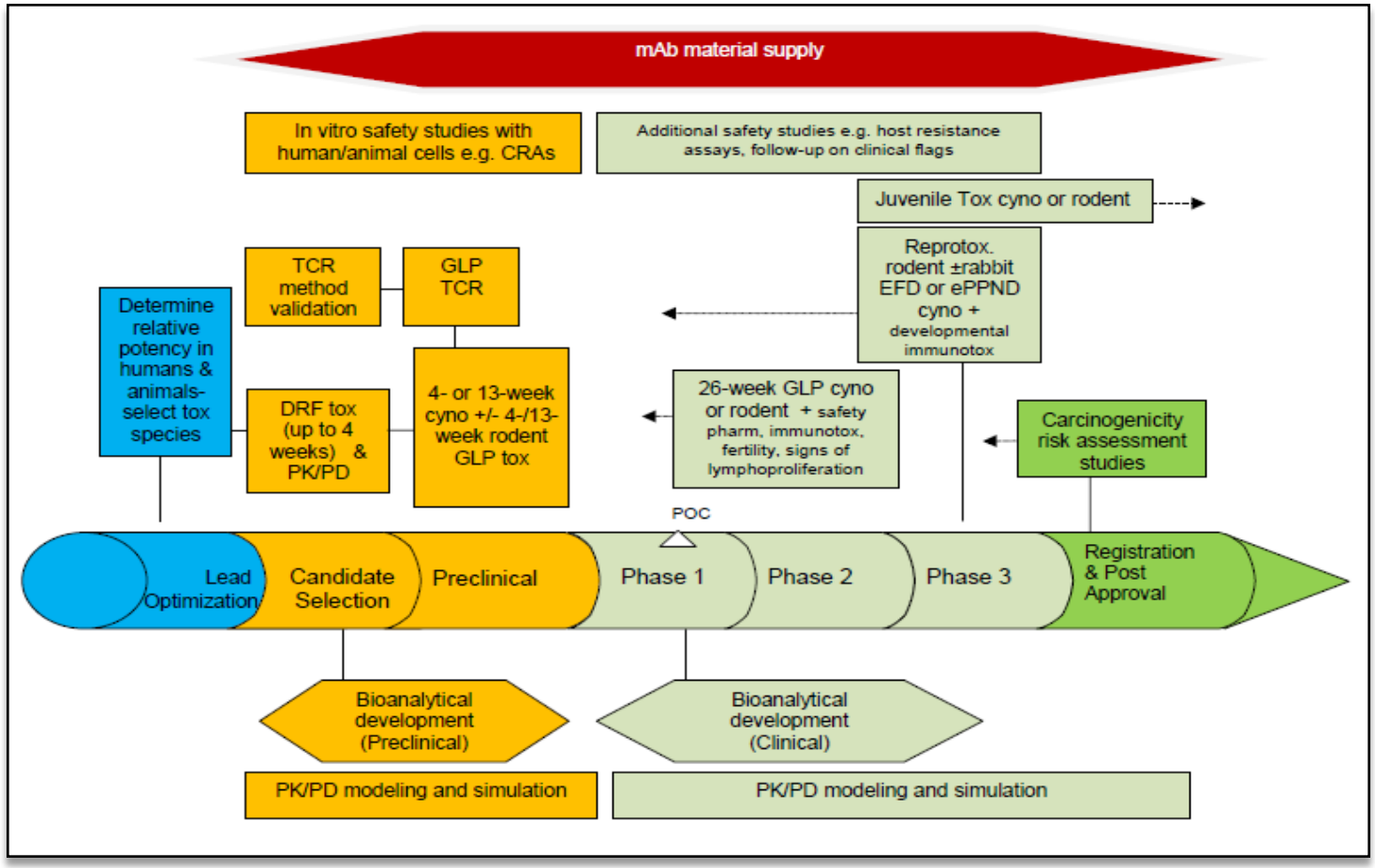
- Development of ADAs
- Increased clearance
- Infusion reactions
- Unclear long term effects for healthy volunteers

Risk Mitigation for Biologics



Optimized Nonclinical Safety Assessment Strategies Supporting Clinical Development of Therapeutic Monoclonal Antibodies Targeting Inflammatory Diseases

Frank R. Brennan,* Annick Cauvin, Jay Tibbitts, and Alison Wolfreys
Preclinical Safety, New Medicines, UCB-Celltech, Slough SL1 3WE, UK



DRUG DEVELOPMENT RESEARCH 75 : 115–161 (2014)

UNIVERSITY OF LIVERPOOL | THE WOLFSON CENTRE FOR PERSONALISED MEDICINE

MRC Centre for Drug Safety Science

Nonclinical strategy considerations for safety pharmacology: evaluation of biopharmaceuticals

Hugo M Vargas†, Hamid R Amouzadeh & Michael J Engwall

- Science based approach
  - Clear rationale, evidence based = regulatory acceptance
- Integrated safety pharmacology
  - Do with GLP tox (usually NHP 4- or 13- week)
  - Less concern for hERG inhibition / interaction
  - Less concern for CNS toxicity (NB indirect effects)
  - Immunogenicity assessment in vivo and in vitro
- Immuno-pharmacology
  - Human / animal species
  - Dose-response
  - Assessment of CRS

Expert Opin. Drug Saf. (2013) 12(1):91-102

TABLE 4. Toxicology Studies Performed for Approved mAbs/Fusion Proteins for Inflammatory Disease Indications

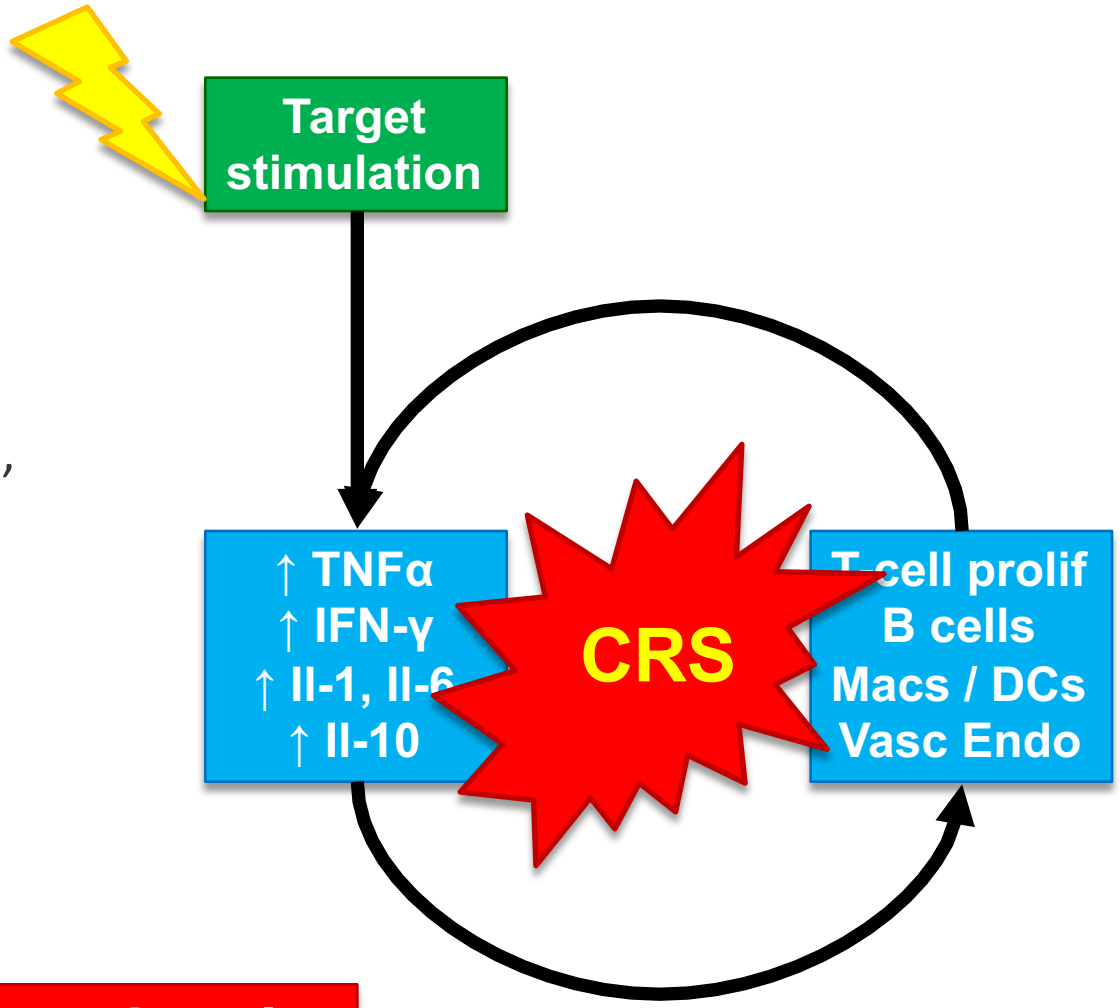
Generic name	Target	Relevant species	Major tox. studies performed	Observed effects
Muromonab	CD3	Rhesus macaque	2-week tox. study with 10-month observation period	↑number and size of germinal centers in spleen and LNs.
Daclizumab	IL-2R	Cynomolgus macaque (cyno)	Single dose tox. in mice and rabbits 4-week tox. in cynos	None
Rituximab	CD20	Cyno	8-week tox., EFD and PPND studies in cynos	↓B cells in blood and lymphoid organs of mothers and infants
Basiliximab	IL-2R	Rhesus, Cyno	4-week tox. in rhesus macaques, EFD study in cynos.	None
Infliximab	TNFα	Mouse (surrogate)	26-week tox., fertility, EFD & PPND studies with anti-mouse TNFα surrogate mAb in mice	None (possibly slight ↓male fertility, bilateral crystalline deposits in lens capsule of males of unknown relevance)
Etanercept	TNFα	Cyno, Rat, rabbit	26-week tox. study in cynos 12-week immunogenicity studies in mice, rats and rabbits; EFD and PPND studies in rats, EFD study in rabbits.	None (mild ↑eosinophil and lymphocytes at injection site)
Alemtuzumab	CD52	Cyno	30-day tox. study in cynos (ADA restricted dosing duration)	↓WBCs, lymphopenia, neutropenia, ↓serum total protein & albumin
Adalimumab	TNFα	Cyno	39-week tox. and EFD-PPND studies in cynos	↓thymus weight and involution, ↓splenic follicular centers and DCs;↓T cells in thymus and B cells in spleen (males only)
Omalizumab	IgE	Cyno	26-week tox.,fertility (mating) & EFD-PPND studies in cynos; juvenile tox. studies of up to 26 weeks in cynos.	↓Platelets in adult cynos, TCP in juvenile cynos
Efalizumab	CD11a (LFA-1)	Mouse (surrogate)	26-week non-terminal study in chimpanzees; 4-week tox., fertility, EFD and PPND studies with anti-mouse CD11a surrogate mAb in mice; 6-month carcinogenicity study in p53 KO mice.	↓humoral response, ↑WBC, ↓LN cellularity; ↓DTH a and TDAR responses
Alefacept	CD2	Cyno, baboon	44–47-week tox. and EFD-PPND studies in cynos; 3-month study in baboons	↓T cells in blood and lymphoid organs; mild↓TDAR to KLH but not HSA; lymphoma in 1 animal after 28 weeks.
Natalizumab	CD49d (VLA-4)	Cyno, rhesus, guinea pig, mouse	2-week tox. study in mice; 26-week tox. and EFD studies in cynos, 26-week juvenile tox. study in cynos. Fertility, EFD and PPND in guinea pigs; 4-week combination tox. study with Avonex (IFN-β) in rhesus; assesed effect of mAb on VLA-4-expressing tumor growth in vitro and in SCID and nude mice	(↑WBC, ↑reticulocytes, ↑spleen weight and follicular hypertrophy, ↓liver and thymus weight (neonates only), lymphoplasmacytic inflammation of the colon; ↓female fertility; glomerulonephritis in cynos related to immunogenicity; anaphylaxis in guinea pigs related to immunogenicity.

Key things to look for

- Comparability with humans
  - Animal model (NHP, homologous target in other?)
  - In vitro work
- Target
  - Nature (cell based, soluble)
  - Receptor occupancy
  - Dose response curve
  - Evidence of CRS or immune depletion
- High risk IMP for CRS
- Immunogenicity

Risk of CRS

- ESG / EMEA
- High risk if:
  - Bind TLRs etc
  - Bind 'master switches'
  - Fcγ functionality
  - Multivalent
  - Cause proliferation / expansion
  - Agonistic activity



May have no clear signal from NHP

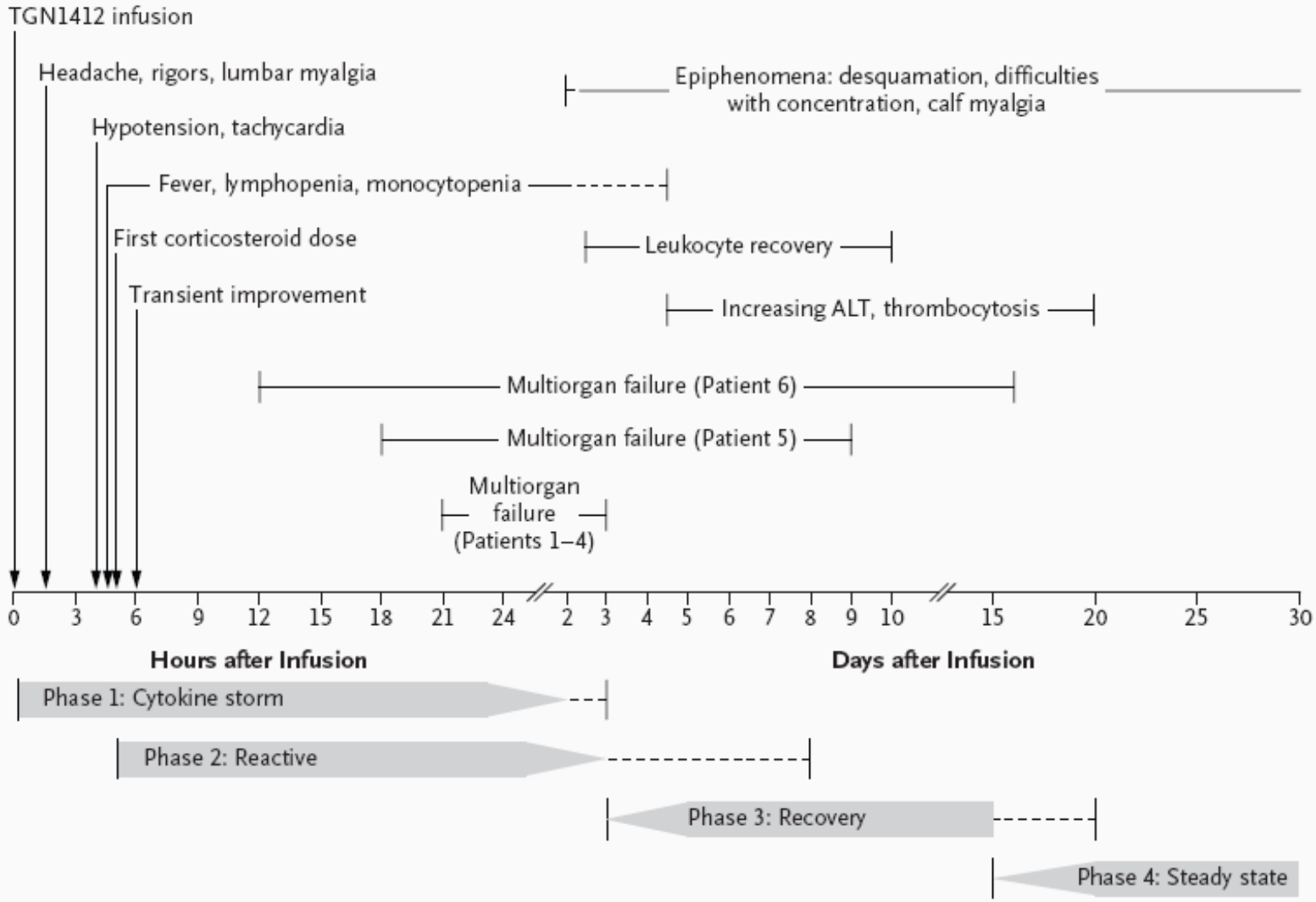


Figure 1. Summary Timeline of the Main Events after Infusion of TGN1412.

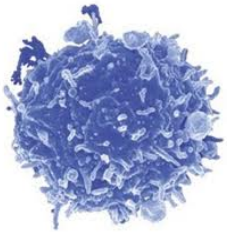
The course is divided into four phases: cytokine storm, reactive, recovery, and steady state. ALT denotes alanine aminotransferase. Dashed lines represent the responses of Patients 5 and 6 (who were the most seriously ill).

NHP vs Human?

CD33 related Siglecs

↓ in man
↑ in CynM

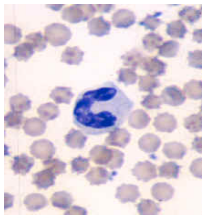
T cell targets

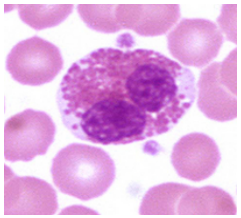


Fc mediated effects

CD16 expression

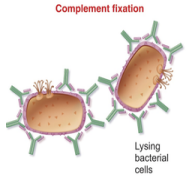
Neutrophil, eosinophil targets





IgG / Compl binding

Different binding effects



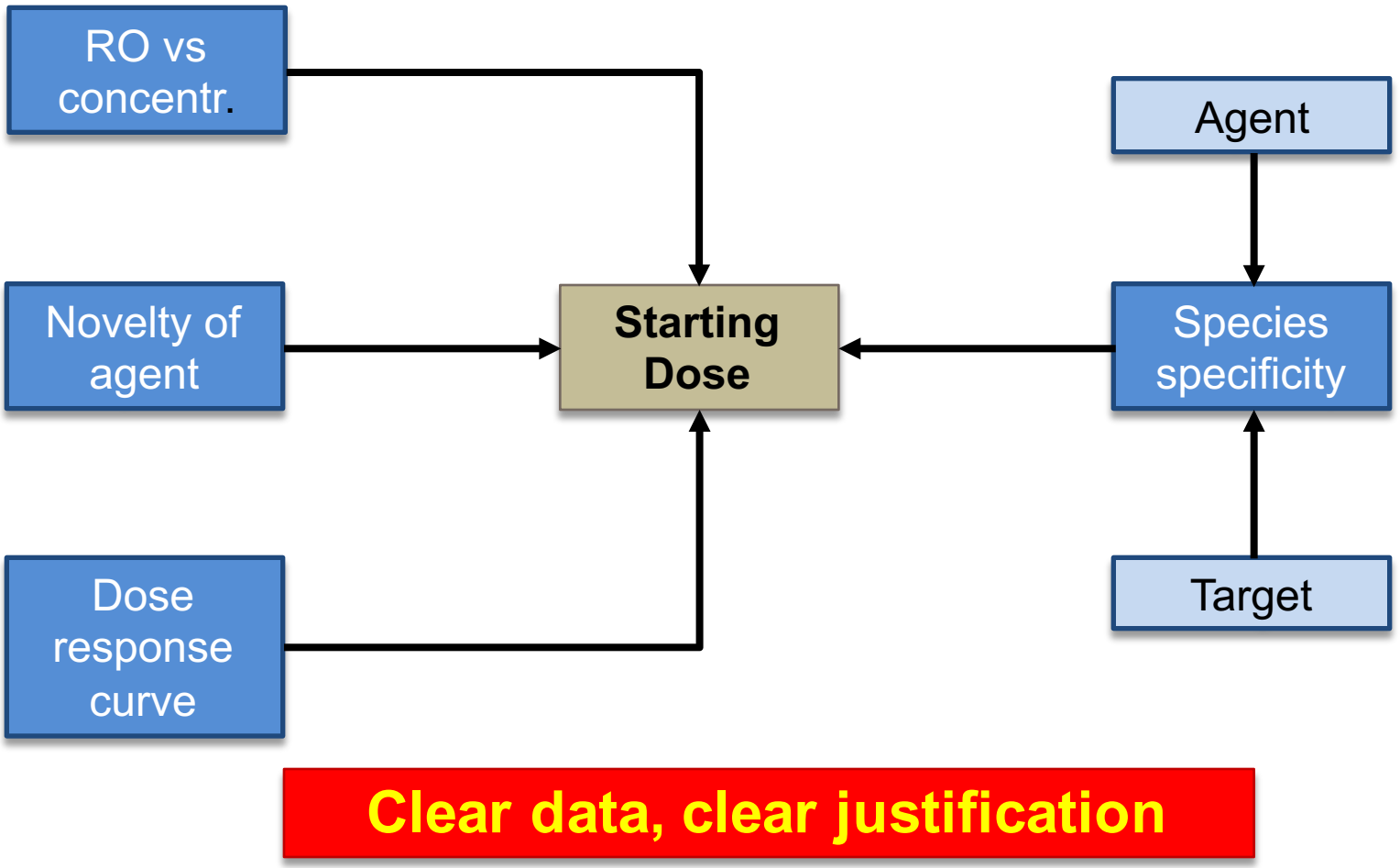
Safety Assessment and Dose Selection for First-in-Human Clinical Trials With Immunomodulatory Monoclonal Antibodies

PY Muller[1] and FR Brennan[2]

- Receptor occupancy model
- Predict starting dose for circa 10% RO
- Beware odd dose response curves
- Utilise a MABEL approach in most cases (always if high risk)
- Consider **all** data that are available

CLINICAL PHARMACOLOGY & THERAPEUTICS | VOLUME 85 NUMBER 3 | MARCH 2009

Considerations for first dose…….



Safety Assessment and Dose Selection for First-in-Human Clinical Trials With Immunomodulatory Monoclonal Antibodies

PY Muller[1] and FR Brennan[2]

$$\text{Eq. 1:}\quad K_D = \frac{[Ab_{free}]\cdot[Ta_{free}]}{[Ab\circ Ta]} = \frac{(Ab_{tot} - Ab\circ Ta)\cdot(Ta_{tot} - Ab\circ Ta)}{Ab\circ Ta}$$

$$\text{Eq. 2:}\quad RO = \frac{Ab\circ Ta}{Ta_{tot}}$$

$$\text{Eq. 3:}\quad Ab_{tot} = \frac{Ab_{molar\ dose}}{V_{initial,plasma}} \Rightarrow Ab_{dose} = Ab_{tot}\cdot MW_{Ab}\cdot V_{initial,plasma}$$

$$\text{Eq. 4:}\quad Ab_{tot} = \frac{RO\cdot Ta_{tot}\cdot\left(RO - 1 - {K_D}/{Ta_{tot}}\right)}{RO - 1}$$

Figure 1 Equations 1–4. Receptor occupancy (RO) for monoclonal antibody/target interaction and calculation of dose. Under equilibrium, the binding of an antibody (Ab) to its target (Ta), leading to the formation of an Ab–target complex (Ab∘Ta), is expressed by the mass–action law outlined in Eq. 1, whereby K_D represents the dissociation constant. In Eq. 2, RO is expressed as the fraction of the Ab–target complex relative to total target expression, TE (Ta_{tot}). In Eq. 3, the dose (Ab_{dose}) of the Ab is expressed by total Ab concentration (Ab_{tot}) multiplied by its molecular weight (MW_{Ab}; assumed to be 150 kDa) and the initial plasma distribution volume after intravenous administration ($V_{initial,plasma}$; assumed to be 0.036 l/kg). By combining Eqs. 1 and 2, the Ab_{tot} value can be expressed as shown in Eq. 4.

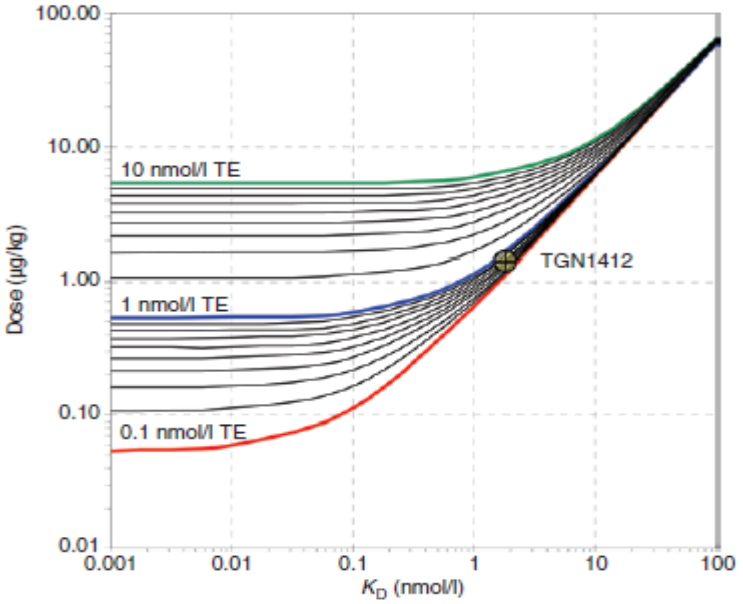
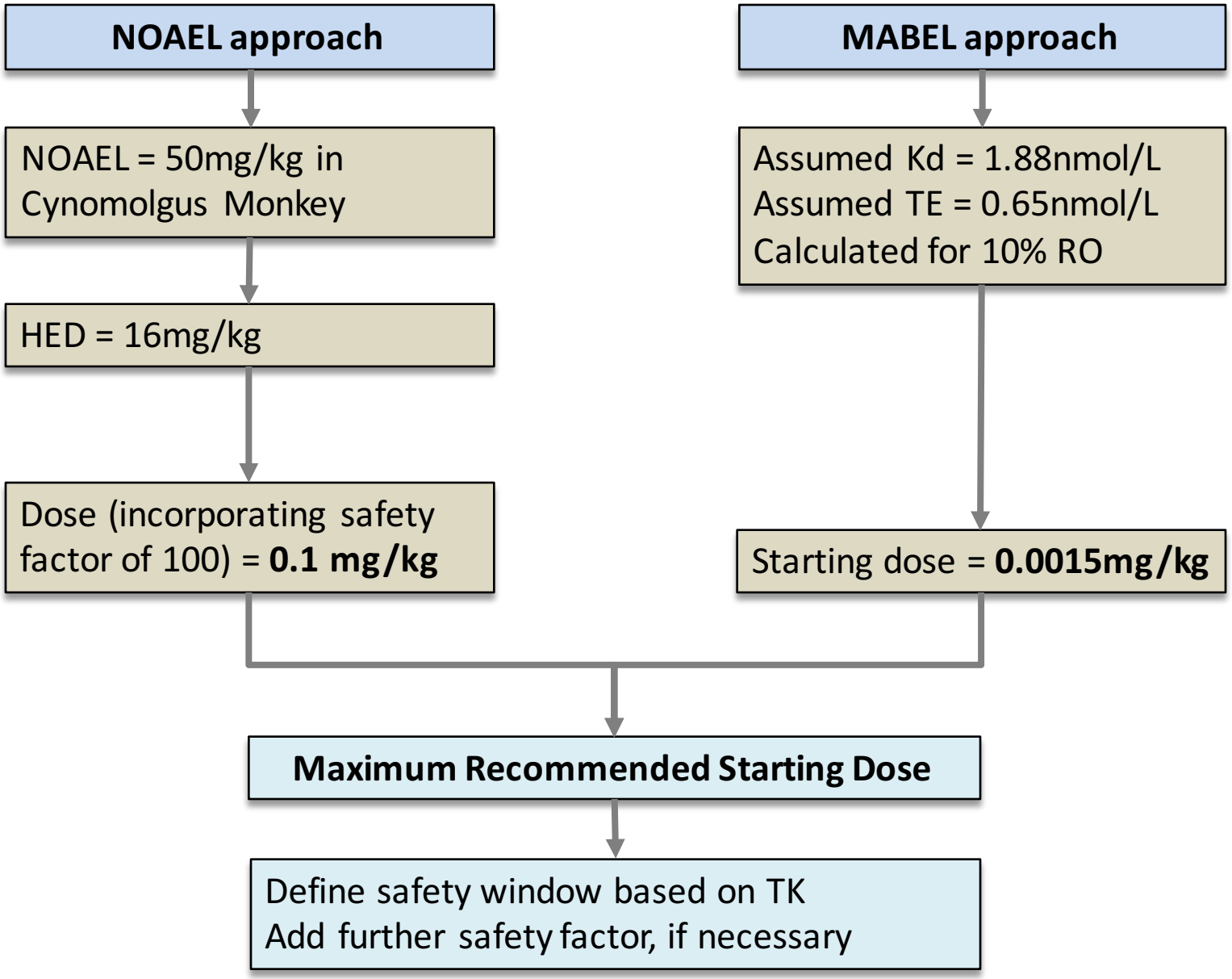


Figure 2 Graphical representation of monoclonal antibody dose leading to 10% receptor occupancy (RO). The antibody dose leading to an RO of 10% is calculated based on Eqs. 3 and 4 in **Figure 1** as a function of the dissociation constant (K_D) for total target expression ranging from 0.1 nmol/l (red line) through 1 nmol/l (blue line) to 10 nmol/l (green line) in 10 scaling steps each. Molecular weight is assumed to be 150 kDa; initial plasma distribution volume is assumed to be 0.036 l/kg. The TGN1412 dose leading to a predicted RO of 10% in humans is depicted.

CLINICAL PHARMACOLOGY & THERAPEUTICS | VOLUME 85 NUMBER 3 | MARCH 2009


NOAEL approach
NOAEL = 50mg/kg in Cynomolgus Monkey
HED = 16mg/kg
Dose (incorporating safety factor of 100) = 0.1 mg/kg
MABEL approach
Assumed Kd = 1.88nmol/L
Assumed TE = 0.65nmol/L
Calculated for 10% RO
Starting dose = 0.0015mg/kg
Maximum Recommended Starting Dose
Define safety window based on TK
Add further safety factor, if necessary

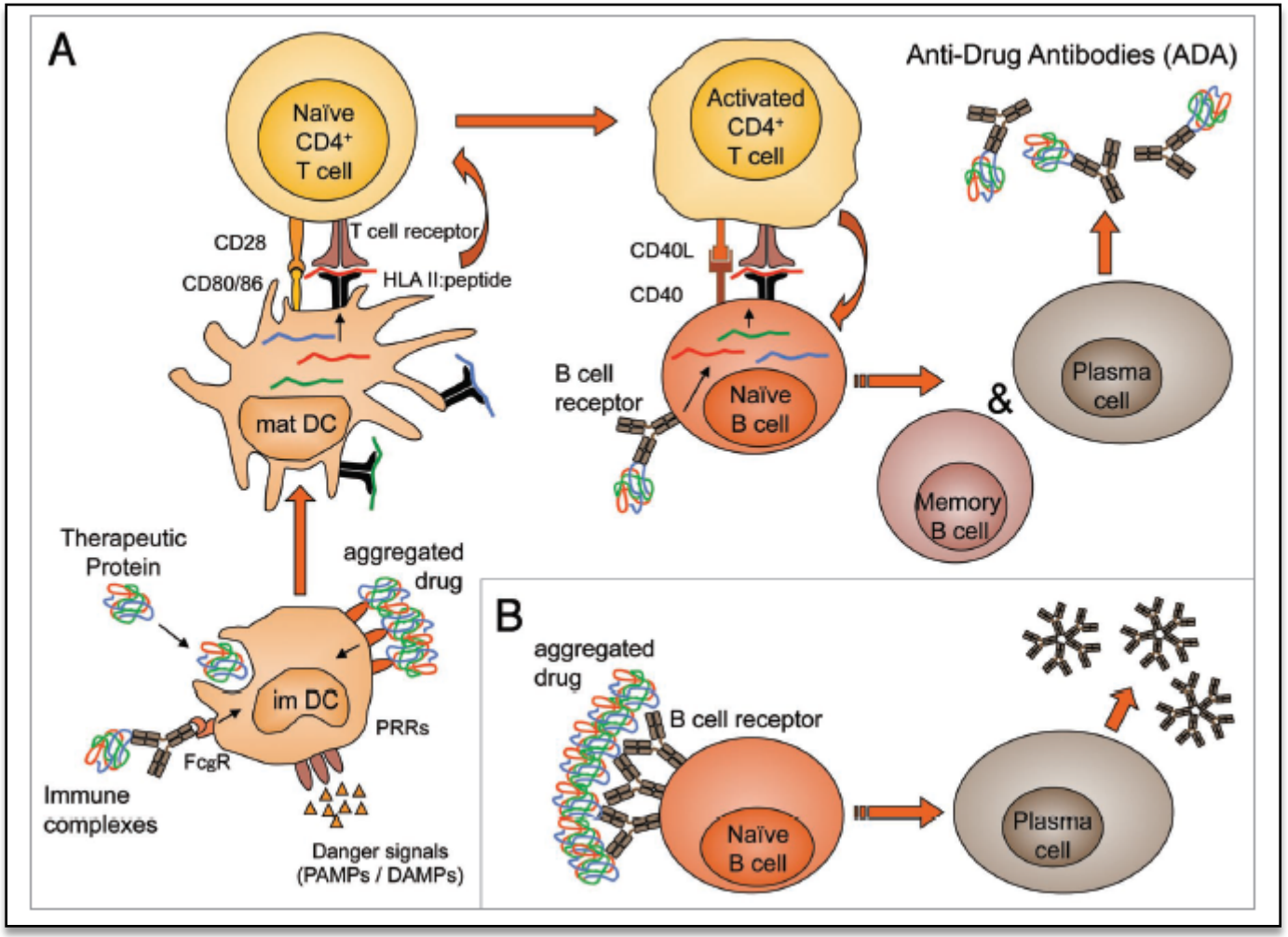
Immunogenicity

- Rational choice of mAb
- Avoid significant complement binding (avoid CDC)
- Activation of immune cells (NK, phagocytes, dendritic cells)
- Depletion of immune cells
- Cleavage of Ab

Table 3. Key functional characteristics of human IgG subclasses

Subclass	IgG1	IgG2	IgG3	IgG4
Serum half-life (days)	21	20	7	21
FcRn[a] binding	++	++	+	++
FcγRI[b] binding	++++	-	++++	+++
FcγRIIA[c] binding	+++	+	++	+
FcγRIIB[d] binding	++	+	++	+
FcγRIIC[e] binding	++	+	++	+
FcγRIIIA[f] binding	+++	+/-	+++	+
FcγRIIIB[g] binding	++	-	++	-
C1q[h] binding	++	+	+++	-

Induction of Anti-Drug Abs



Cross-reactive and pre-existing antibodies to therapeutic antibodies—Effects on treatment and immunogenicity

Karin A van Schie[1], Gerrit-Jan Wolbink[1,2], and Theo Rispens[1,*]

- Specific to mAb
  - Anti-idiotype
- Cross-reactive / pre-existing
  - Rheumatoid factor
  - Anti-allotype
  - Anti-hinge
  - Anti-glycan

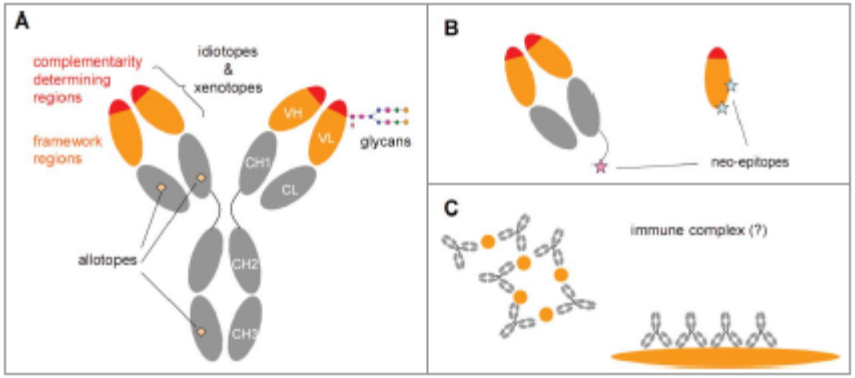


Table 1. Reports on consequences of pre-existing antibodies

Drug	Effect
abciximab	presence of anti-hinge antibodies might be correlated with thrombocytopenia
cetuximab	IgE antibodies recognizing alpha-gal sugar moieties caused anaphylactic reactions in patients from certain regions in the United states
GSK1995057	in vitro and in vivo cytokine release was associated with antibodies recognizing framework regions of the drug (a VH domain) but not the intact antibody
rituximab	decreased in vitro complement-dependent cytotoxicity of rituximab was observed in the presence of rheumatoid factor

mAbs 7:4, 662–671; July/August 2015;

Pre-Existing Biotherapeutic-Reactive Antibodies: Survey Results Within the American Association of Pharmaceutical Scientists

Li Xue,[1,7] Michele Fiscella,[2] Manoj Rajadhyaksha,[3] Jaya Goyal,[4] Claire Holland,[5] Boris Gorovits,[1] and Alyssa Morimoto[6]

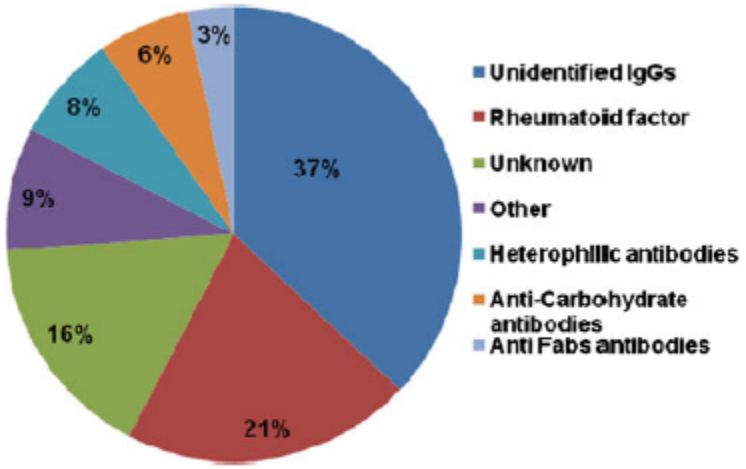
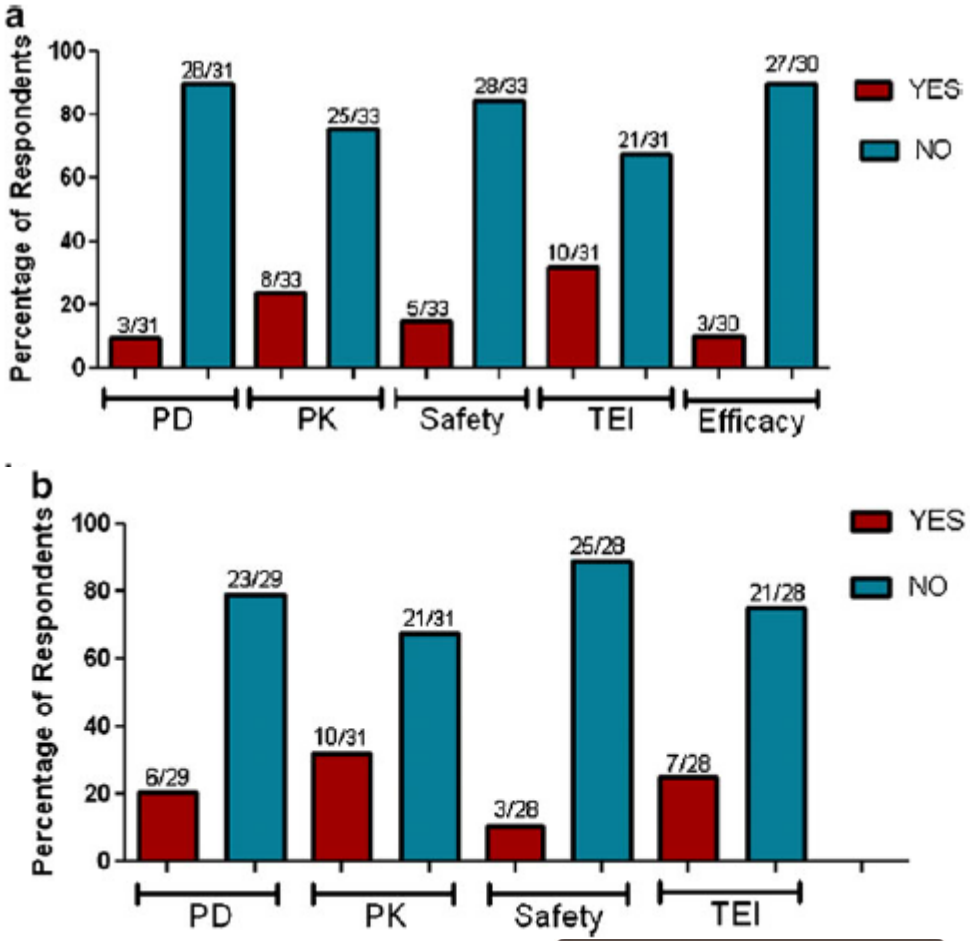


Fig. 2. Specificity of pre-existing antibodies identified in clinical samples



The AAPS Journal, Vol. 15, No. 3, July 2013

UNIVERSITY OF LIVERPOOL | THE WOLFSON CENTRE FOR PERSONALISED MEDICINE

MRC Centre for Drug Safety Science

Evaluation of Pre-existing Antibody Presence as a Risk Factor for Posttreatment Anti-drug Antibody Induction: Analysis of Human Clinical Study Data for Multiple Biotherapeutics

Li Xue[1,2] and Bonita Rup[1]

Table III. Prevalence of Pre-existing Antibodies from All Studies

	Percentage	Number of subjects
In all study subjects	5.6%	(103/1830)
In healthy volunteers	0.6%	(3/499)
In all disease populations	7.5%	(100/1331)
In disease populations excluding RA	4.2%	(38/911)
In RA patients	14.8%	(62/420)

Table IV. Prevalence of Pre-existing Abs from Studies Associated with Pre-existing Antibodies

	Percentage	Number of Subjects
In all study subjects	10.8%	(103/950)
In healthy volunteers	3.6%	(3/84)
In all disease populations	11.5%	(100/866)
In disease populations excluding RA	8.5%	(38/446)
In RA patients	14.8%	(62/420)

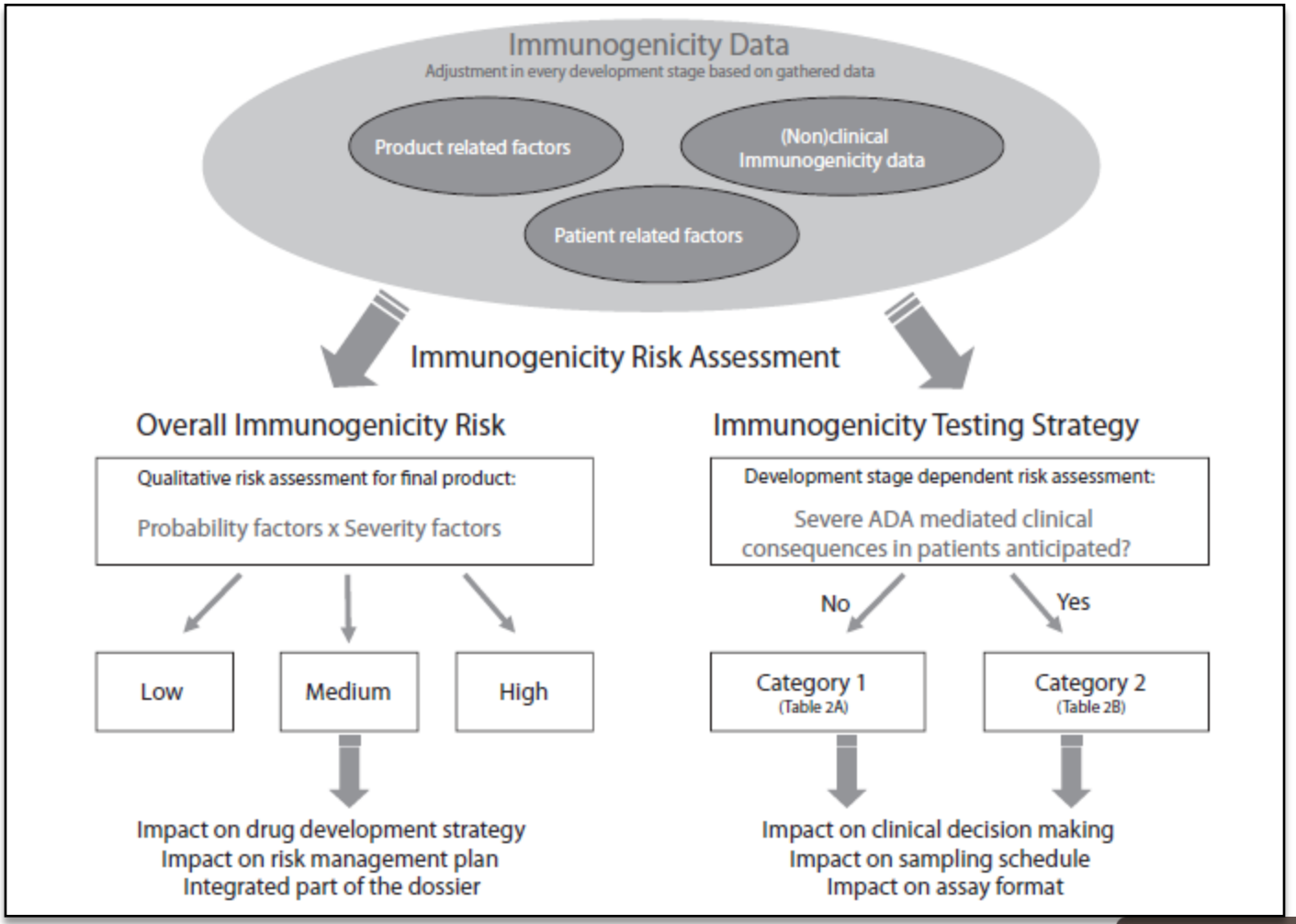
Table V. Association with Posttreatment ADA Induction at Product Level

	Association with ADA induction	No association with ADA induction
Products with Pre-Ab	67% (4/6[a])	33% (2/6)
Products without Pre-Ab	60% (3/5)	40% (2/5)
Products with Pre-Ab and studied in RA	100% (3/3)	0% (0/3)

[a] One of the seven products associated with pre-existing antibodies (Pre-Ab) was excluded from comparison with posttreatment ADA induction, as the pre-Ab was only observed in placebo group

A fit-for-purpose strategy for the risk-based immunogenicity testing of biotherapeutics: a European industry perspective

Cathelijne Kloks [a,1], Claudia Berger [b,1], Pierre Cortez [c], Yann Dean [d], Julia Heinrich [e], Lisbeth Bjerring Jensen [f], Vera Koppenburg [g], Stefan Kostense [h], Daniel Kramer [i,*], Sebastian Spindeldreher [j], Hishani Kirby [k]



A fit-for-purpose strategy for the risk-based immunogenicity testing of biotherapeutics: a European industry perspective

Cathelijne Kloks [a,1], Claudia Berger [b,1], Pierre Cortez [c], Yann Dean [d], Julia Heinrich [e], Lisbeth Bjerring Jensen [f], Vera Koppenburg [g], Stefan Kostense [h], Daniel Kramer [i,*], Sebastian Spindeldreher [j], Hishani Kirby [k]

Table 2
Essential immunogenicity testing strategy for biotherapeutics; category 1 (panel A) and category 2 (panel B).

	Non-clinical	Phase ISingle dose	Phase IMultiple dosing	Phase II and III
ADA assay format	Screen (99.9th)	Screen + conf	Screen + conf	Screen + conf
Sample collection	Frequent	At least baseline and end-of-study samples	Frequent	Frequent
Samples to be tested	Event driven	Event driven	At least baseline and end-of-study	At least baseline and end-of-study
Execution of testing	Batch wise at end of study if required	Batch wise at end of study if required	Batch wise at end of study	Batch wise at end of study
Neutralization	–	–	–	PD/If of added value: CLB or CBA
Characterization	–	–	If of added value	If of added value

B Testing strategy category 2. Biotherapeutics with expected potential to elicit ADA-mediated severe clinical consequences

	Non-clinical	Phase ISingle dose	Phase IMultiple dosing	Phase II and III
ADA assay format	Screen (99.9th)	Screen + conf	Screen + conf	Screen + conf
Sample collection	Frequent	At least baseline and end-of-study samples	Frequent	Frequent
Samples to be tested	Event driven	All samples	All samples	All samples
Execution of testing	Batch wise at end of study	Batch wise for each cohort or at end of study for single dose	In a timely manner	In a timely manner
Neutralization	If of added value: PD/CLB or CBA	PD/CLB or CBA	PD/CLB or CBA	PD/CLB or CBA
Characterization	–	If of added value; select relevant assays (see Box 4)	If of added value; select relevant assays (see Box 4)	If of added value; select relevant assays (see Box 4)

Quantifying the risks of non-oncology phase I research in healthy volunteers: meta-analysis of phase I studies

Ezekiel J Emanuel,[1,2] Gabriella Bedarida,[3] Kristy Macci,[3] Nicole B Gabler,[4] Annette Rid,[5] David Wendler[6]

Table 2 | Characteristics of drugs used in non-oncology phase I research studies

Characteristics	No (%) of participants receiving study drug by test site location			Total (% are for column)
	USA	Belgium	Singapore	
Previously FDA approved agent				
Total	1143 (44.8)	460 (18.0)	949 (37.2)	2552 (23.1)
Type of agent:				
Small molecule	1143 (44.8)	460 (18.0)	949 (37.2)	2552
Biologic	0 (0)	0 (0)	0 (0)	0
Primary treatment area:				
Neurological/psychiatric	449 (71.6)	116 (18.5)	62 (9.9)	627
Cardiovascular	316 (48.7)	0 (0)	333 (51.3)	649
Pulmonary	0 (0)	0 (0)	202 (100)	202
Gastrointestinal	0 (0)	0 (0)	0 (0)	0
Gynecological	52 (32.5)	60 (37.5)	48 (30.0)	160
Rheumatological	90 (30.1)	149 (49.8)	60 (20.1)	299
Infectious disease	122 (34.3)	77 (21.6)	157 (44.1)	356
Other	114 (44.0)	58 (22.4)	87 (33.6)	259
Investigational agent				
Total	2361 (33.5)	2988 (42.4)	1700 (24.1)	7049 (63.9)
Type of agent:				
Small molecule	2234(33.3)	2782 (41.4)	1700 (25.4)	6716
Biologic	127 (38.1)	206 (61.9)	0 (0)	333
Primary treatment area:				
Neurological/psychiatric	616 (40.2)	547 (35.7)	370 (24.1)	1533
Cardiovascular	830 (62.4)	353 (26.5)	148 (11.1)	1331
Pulmonary	0 (0)	369 (58.2)	265 (41.8)	634
Gastrointestinal	0 (0)	207 (55.6)	165 (44.4)	372
Gynecological	40 (13.4)	180 (60.2)	79 (26.4)	299
Rheumatological	272 (35.1)	207 (26.7)	296 (38.2)	775
Infectious disease	355 (35.1)	525 (52.0)	130 (12.9)	1010
Other	248 (22.6)	600 (54.8)	247 (22.6)	1095

Table 3 | Frequency and severity of adverse events in non-oncology phase I research studies

Characteristic of studies	Adverse events			Total (% are for column)
	Mild	Moderate	Severe	
No (%) of adverse events				
Total	20 840 (84.6)	3548 (14.4)	255 (1.0)	24 643 (100)
Type of agent:				
Small molecule	20 191 (85.2)	3260 (13.8)	243 (1.0)	23 694 (96.1)
Biologic	649 (68.4)	288 (30.3)	12 (1.3)	949 (3.9)

Table 5 | Serious adverse events in non-oncology phase I research studies

Characteristic of studies	No of serious adverse events by agent type		Total No
	Small molecule	Biologic	
Total	33	1	34
Caused by study drug*	10	1	11
First day of study	4	0	4
Placebo group	4	0	4
Death	0	0	0
Life threatening event	0	0	0
Extended stay in phase I unit or hospital admission	26	1	27
Medical or surgical intervention†‡	11	1	12
Persistent or major disability or incapacity	0	0	0
Birth defect or anomaly	0	0	0

BMJ 2015;350:h3271

Giving monoclonal antibodies to healthy volunteers in phase 1 trials: is it safe?

Elizabeth Tranter, Gary Peters, Malcolm Boyce & Steve Warrington

Hammersmith Medicines Research, London, UK

- Published data are poor
- 36 trials, 1799 HVs
- If TGN1412 included – 1:425 -1:1700 volunteer trials of life-threatening AE
- If TGN1412 excluded – 1:100000-1:1000000

1 A 31-year-old man had received a depot intramuscular injection of flupenthixol, 1 day before he received an intravenous infusion of a new anti-arrhythmic drug (eproxindine) which caused asystole and death [25].

2 A 19-year-old woman committed suicide (by hanging) while she was an inpatient in a phase 1 research unit, shortly after the protocol-specified, abrupt withdrawal of repeat dose treatment with the antidepressant duloxetine [26]. Although suicidal ideation and withdrawal effects of duloxetine have been reported, and young women very rarely hang themselves, the role of duloxetine withdrawal in this tragic event nevertheless remains uncertain.

3 A 24-year-old woman developed ARDS and died from multi-organ failure, after inhaling high dose hexamethonium in an exploratory, 'proof of principle' trial [27]. Although hexamethonium is an 'old' drug, the novel route and high dose mean that the drug was truly an IMP.

Br J Clin Pharmacol / **76**:2 / 164–172

Healthy volunteer or patient?

- An age-old debate!
- Specifically for biologics:
  - Robustness?
  - Immunogenicity?
  - Potential indication?
  - PK?
  - Target expression?
  - Risk of AE vs benefit of treatment?

Workshop report

Nonclinical safety testing of biopharmaceuticals – Addressing current challenges of these novel and emerging therapies

Frank R. Brennan [a,*], Andreas Baumann [b], Guenter Blaich [c], Lolke de Haan [d], Rajni Fagg [e], Andrea Kiessling [f], Sven Kronenberg [g], Mathias Locher [h], Mark Milton [i], Jay Tibbitts [a], Peter Ulrich [f], Lucinda Weir [e]

- Bi-specific scaffolds
- PK/PD designs to optimise safety
- Prediction of cytokine release and CRS
- Better immunogenicity assessment

Key issues

- **Preclinical work**
  - Ensure fit for purpose, relevant to molecule and species
  - Adequate prediction of transition to humans
  - No set design – justify on scientific rationale
- **Transition to clinical studies**
  - Safe starting dose
  - Base on RO and MABEL
  - Interrogation of preclinical work
- **Immunogenicity**
  - Understand impact
  - Ensure that study design incorporates risk of immunogenicity
  - ADA assessment judiciously
- **Trial Participant**
  - Healthies vs patients
  - Previous exposure to biologics?

Concluding thoughts…….

- Predicting risk is key
- Aside from TGN1412, safety profile for biologics are reasonable
- Check ADA levels (as an exclusion) only when risk of immunogenicity is high or in subjects previously exposed to a biologic
- Re-exposure to a biologic is okay, providing risk of immunogenicity is low (to previous and current molecule)

Human regulatory T cells are selectively activated by low-dose application of the CD28 superagonist TGN1412/TAB08

Paula Tabares[1], Susanne Berr[1], Paula S. Römer[1,2], Sergej Chuvpilo[2], Alexey A. Matskevich[3], Dmitry Tyrsin[3], Yury Fedotov[3], Hermann Einsele[4], Hans-Peter Tony[4] and Thomas Hünig[1]

- 3 healthy male volunteers
- Dosed with 5% of original dose over 4-12h
- No significant AEs
- No evidence of CRS

Eur. J. Immunol. 2014. 44: 1225–1236