A Modular MoBi Implementation of Bispecific T-Cell Engager Target-Mediated Drug Disposition: One Base QSP Model for In Vitro, Minimal PBPK and Whole-Body PBPK Simulations

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AGENDA



01. Introduction

02. State of the art for modeling

03. One solution for multiple purpose

04. Conclusions & Next Steps



1. Introduction

Bispecific Antibodies: A Long-Standing Therapeutic

Bispecific antibodies combine two binding specificities in one molecule—and the concept is not new. “Hybrid antibodies” with dual specificity were already described in the 1960s.

The concept

Univalent fragments of two specificities recombine into a bivalent hybrid.

Recombination of a Mixture of Univalent Antibody Fragments of Different Specificity[1]

A. NISONOFF
M. M. RIVERS

Department of Microbiology
University of Illinois
Urbana, Illinois

Received March 27, 1961

oxidized, the recombined product contains a considerable amount of bivalent 5S "hybrid antibody" (6, 7).[1] The recombination of fragments of two different specificities appears to be essentially random, as shown by quantitative analysis of the

Nisonoff & Rivers (1961). [DOI: 10.1016/0003-9861(61)90296-x]

The proof

One hybrid pulls two distinct red-cell types — each bearing a different antigen.

SEROLOGIC DEMONSTRATION OF DUAL SPECIFICITY OF RABBIT BIVALENT HYBRID ANTIBODY*

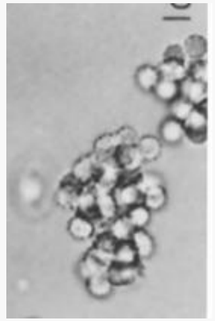
BY H. H. FUDENBERG, M.D., GENEVIEVE DREWS, M.D., AND A. NISONOFF, PH.D.

(From the Department of Medicine, University of California School of Medicine, San Francisco, and the Department of Microbiology, University of Illinois, Urbana)

PLATE 5

(Received for publication, September 4, 1963)

sults indicate that a reconstituted 5S antibody can induce passive hemagglutination, and that reconstituted 5S hybrid antibody preparations produce passive mixed agglutination of two visually distinguishable red cell types, each coated with a different antigen. With an artificial mixture of 5S antibodies (not hybridized), mixed agglutination does not occur; the two cell types are aggluti-



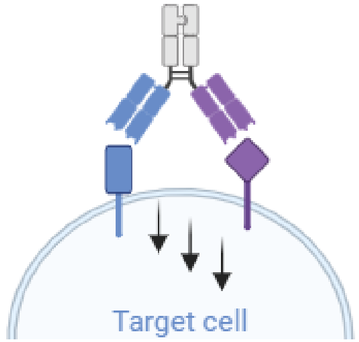
Obtained by peptic digestion (pepsin plus mild reduction of purified antibody) and then recombined by oxidation

Fudenberg, Drews & Nisonoff (1964). [DOI: 10.1084/jem.119.1.151]

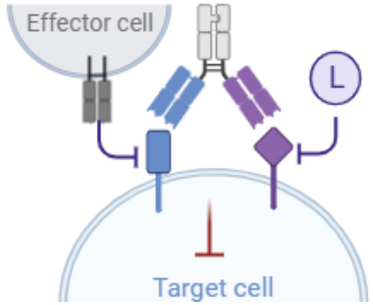
Fast-forward to today — what changed isn't the concept, but our ability/technology to engineer it

Bispecifics Go Beyond T-Cell Engagement

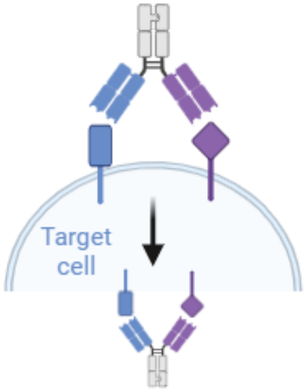
Mutagenesis and purification advances have produced a "zoo“/variety of bispecific antibodies and fragments — varied not just in shape but also in function



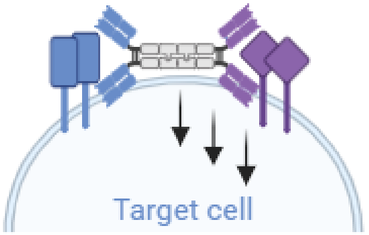
1. Receptor activation



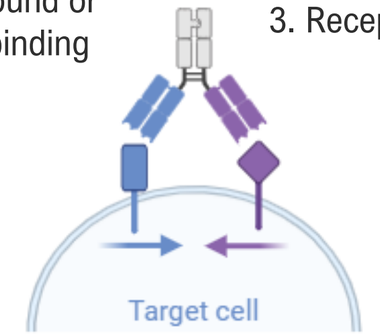
2. Receptor blocking and inhibition cell-bound or soluble ligand binding



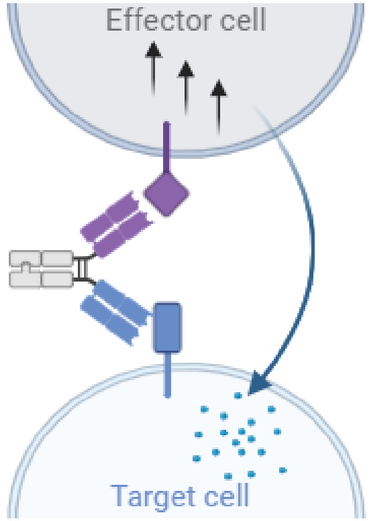
3. Receptor internalization



4. Receptor clustering



5. Receptor association



6. Bicellular binding and retargeting of effector cells

A bispecific T-cell engager is a protein that simultaneously binds through a target antigen on a tumor cell and CD3 on a T-cell to form a TCR-independent artificial immune synapse and circumvent HLA restriction

Vafa & Trinklein (2020) [DOI: 10.3389/fonc.2020.00446]

Adopted from Dickopf, Georges & Brinkmann (2020) [DOI: 10.1016/j.csbj.2020.05.006]
Created with Biorender.com

Bispecific TCEs: where can QSP help?

A large, diverse pipeline brings complex challenges — many of which can be addressed with modeling, especially QSP.

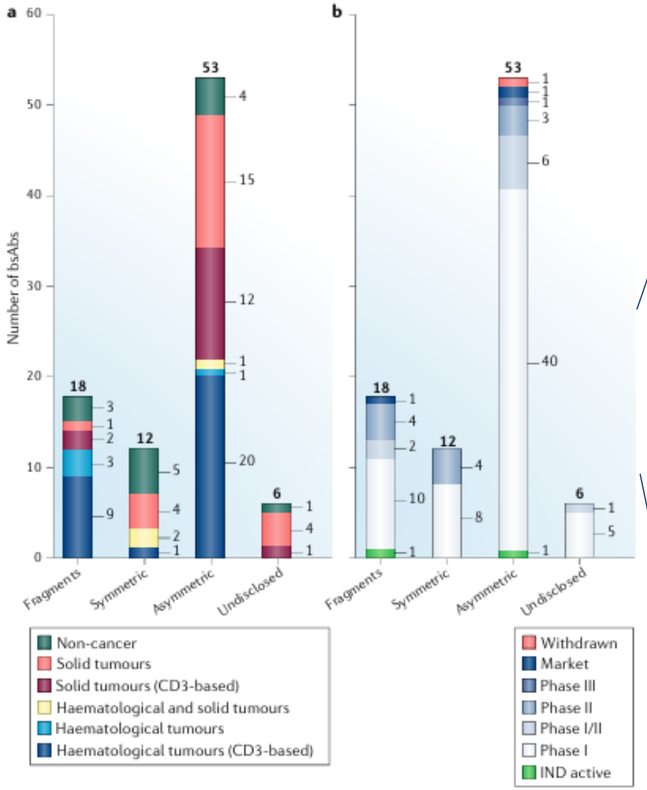


Fig. 3 | **Overview of the clinical development pipeline for bsAbs.** The agents are classified by format class and divided by indication and CD3-mediated concepts (T cell engagement) in panel **a** and development stage in panel **b**. bsAb, bispecific antibody; IND, investigational new drug.

The 2019 clinical pipeline: ~85 bsAbs by format, indication and stage. Extracted from Labrijn et al. (2019), [DOI:10.1038/s41573-019-0028-1]

	Key questions	Challenges	The QSP layer
DISCOVERY	Antibody design, epitope and affinities	Multiple formats and design options	Format and affinity – property relationships
PRECLINICAL	MABEL Starting Dose Regulatory precedence	Context dependency of assays	In vitro in vivo extrapolation
PHASE 1	Dose escalation and optimization Regulatory precedence	Dose-limiting toxicities and patient variability; large gap to effective dose	Dose – exposure – effect relationships
PHASE 2	Therapeutic-window expansion	Narrow therapeutic window; responder heterogeneity; complex regimens and combinations	Responder profiling, digital twins biomarkers etc
LATER STAGES	Long-term outcomes and approval	Durable response and long-term tolerability; risk/benefit for labeling; special populations; cytokine-mediated DDI	IL-6 DDI; dosing in special populations

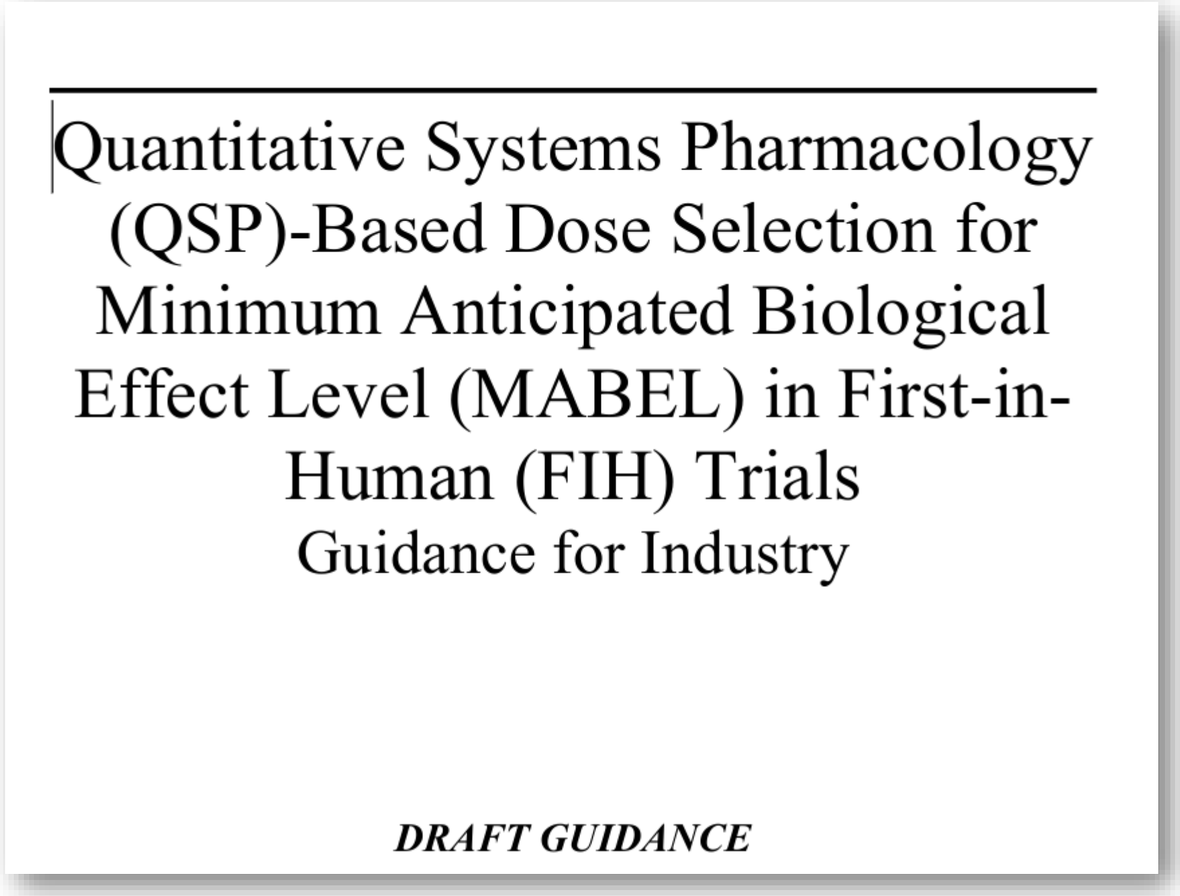
Ball et al. (2023) [DOI:10.1080/19420862.2023.2181016]
Qi, Liao & Cao (2023) [DOI:10.1016/j.tips.2023.09.009]

Bispecific TCEs: where can QSP help?

Regulatory precedence and new MABEL draft guidance

1 – First in Human Trial design

- **Use relevant experiments** as preclinical base material:
  binding in human-derived cells or tissue for human-specific targets.
- **Model** the pharmacology mechanistically:
  PK → target engagement → signalling → biological response
- **Specify** target expression, turnover, protein binding and site of localisation as explicit inputs.
- **Parameterize** species differences — physiology, receptor density, PD response — before translating animal data.
- **Propagate** human biological variability into the predictions.
- **Test** alternative model structures, not just parameter sensitivity; declare omitted biology.
- **Verify** the implementation — no dose → no effect; high dose → saturation; stable stochastic variability.
- **Simulate** doses around the candidate MABEL to expose dose-response steepness.
- **Default** to the lowest estimate when QSP and conventional MABEL disagree, unless retrospectively validated on a same-MOA compound.
- **Justify** the trimer/synapse formulation and cytokine readout — the guidance names T-cell activation and cytokine release as the risk case but prescribes no structure.

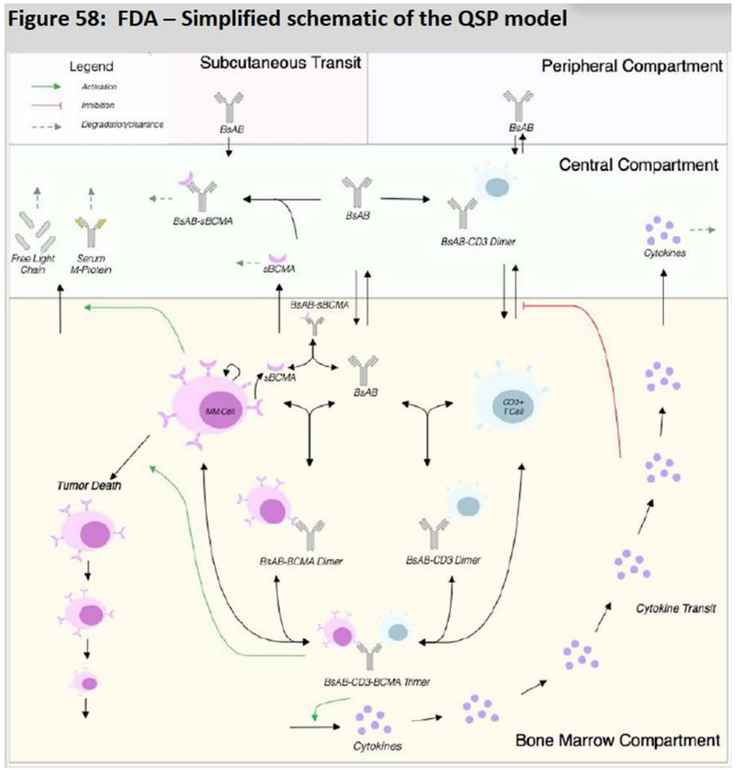


Bispecific TCEs: where can QSP help?

Regulatory precedence and new MABEL draft guidance

2 – Clinical trial dosing decisions

Regulatory use case: Elranatamab BCMAxCD3 in multiple Myeloma
www.accessdata.fda.gov/drugsatfda_docs/nda/2023/761345Orig1s000MultidisciplineR.pdf



Explore the predicted efficacy at different dosing regimens in high/low baseline soluble BCMA populations, compare the maintenance of responses with and without transition from once a week (QW) to twice a week (Q2W) after 24 weeks for participants with persistent responses

Deliverables

- **Executable scripts**, "well documented, with each line/block of scripts followed by relevant descriptions and notes"
- **Datafiles** — input, intermediate and output
- **Model equation files** — complete list of ODEs and all other equations
- **Parameter definition files** — description, value, **units**, sources, parameter-specific assumptions
- **Data utility file** — all data sources informing parameters at each modeling stage
- **Model assumption files** — complete list with justifications
- **Hardware definition files** — hardware specs, OS, all software and tools to reproduce the simulations

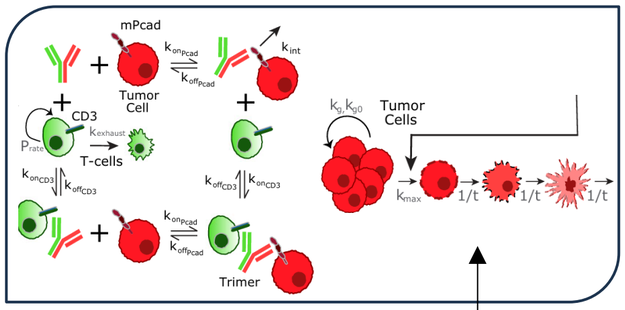


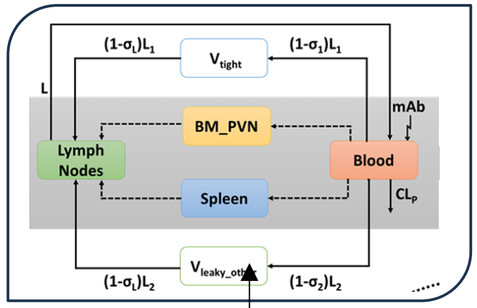
2. State of the art for modeling

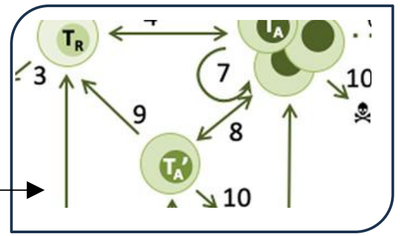
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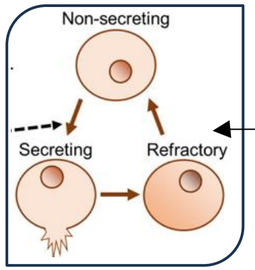
The bsTCE QSP modeling landscape

Published bsTCE models often share set of common mechanisms — i.e. ternary complex formation, tissue distribution, T-cell activation, tumor killing, cytokine release **— but tend to not converge to one setup/differ in structure,** each model is tailored to the specific question being addressed.









	2018 Schropp et al.	2019 Betts et al.	2020 Jiang et al.	2020 Hosseini et al.	2021 De Vyver et al.	2022 Yoneyama et al.	2023 Flowers et al.	2023 Wedell et al.	2024 Liao et al.
COMPARTMENTS	○	○	+	+	-	++	-	○	+
RECEPTORS	++	+	○	-	○	+	++	○	+
T-CELLS	-	○	-	++	++	○	-	(-)	++
CYTOKINES	-	-	○	○	-	○	-	++	-

Whole body PBPK for T-cell engagers?

Scope and tooling

Site of action exposure predictions: T-cell crosslinking in different tissues driving efficacy and safety

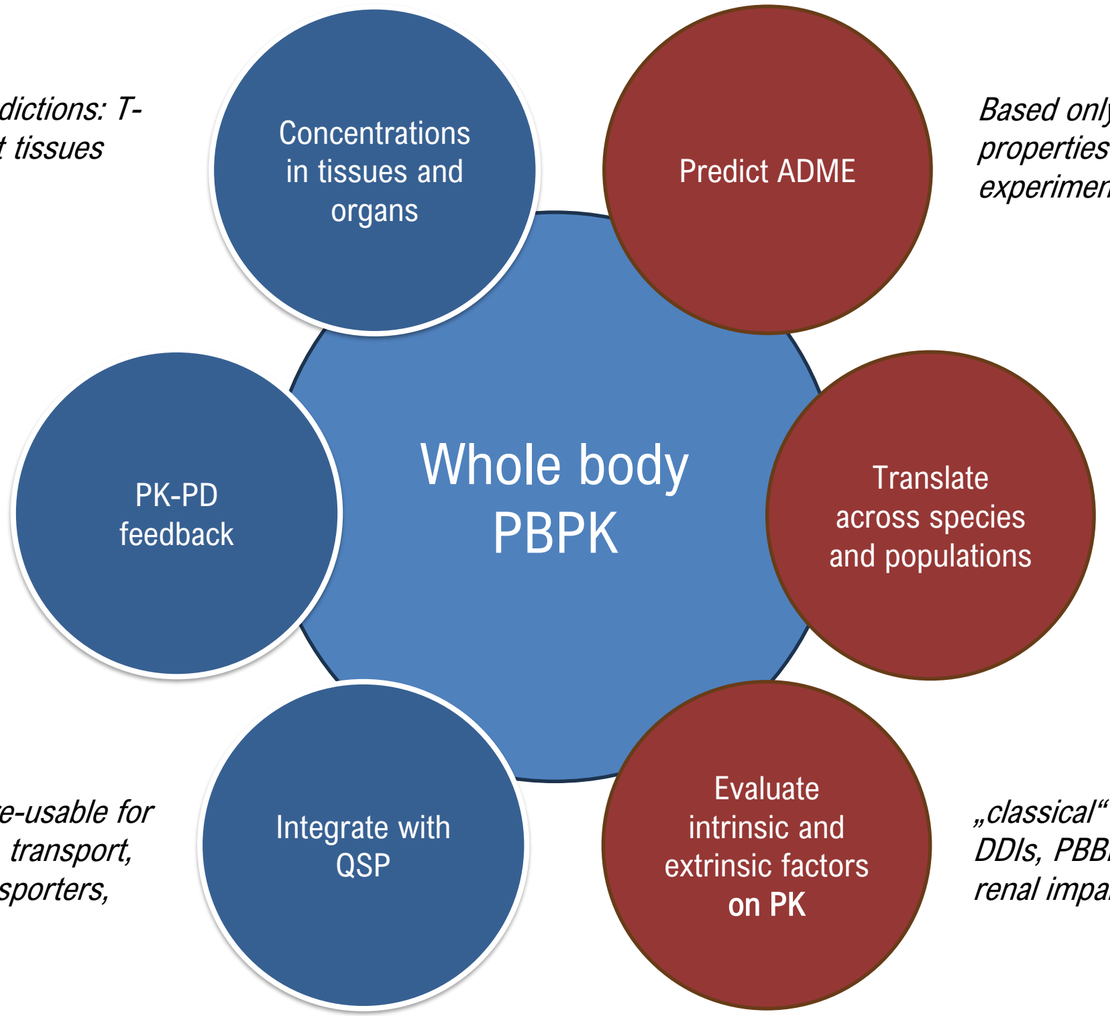
Based only on molecule (structure) properties + in vitro ADME-relevant experiments and known biology

Mechanisms can link effects back to ADME (antigen sinks, cell depletion / proliferation, inflammation)

Predefined species‘ system parametrizations and population generation

„Infrastructure“ re-usable for QSP: expression, transport, metabolism, transporters, ***binding***

„classical“ PBPK Contexts of Use: DDIs, PBBM, hepatic impairment, renal impairment, disease



See Qin (2024)
10.1016/j.apsb.2024.03.027
Scheme adopted from:
Gibbs et al. (2020) 10.1002/jcph.1706

Whole body PBPK for T-cell engagers?

Practical Applicability: Implemented T cell PBPK model (Khot et al. 2019) into PK-Sim protein mouse PBPK

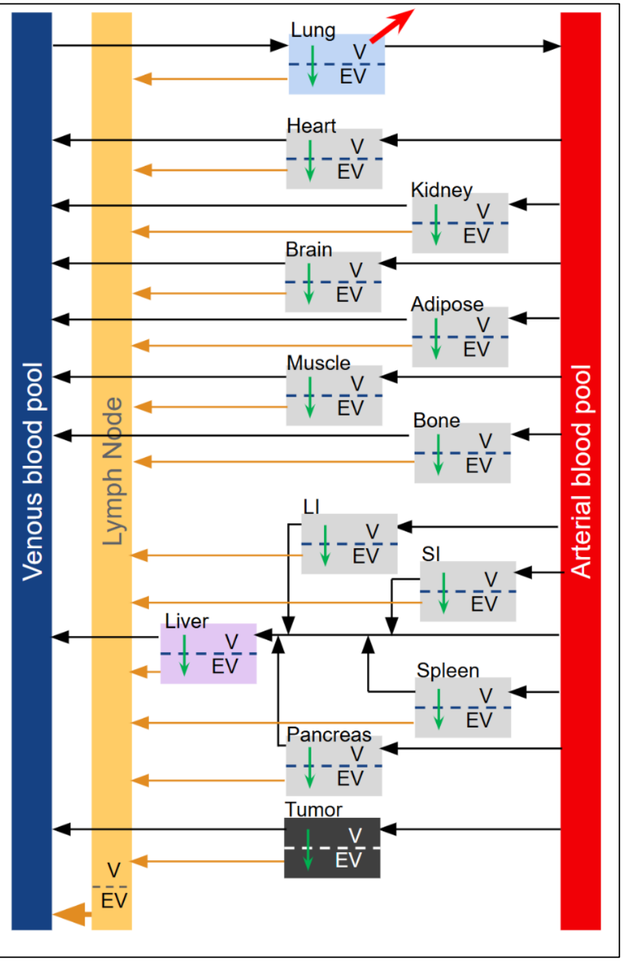
1. Add/Implement literature based T cell module

2. Add/modify Single location-agnostic lymph node compartment

3. Adjust PKSim parameters to fit gD antibody
- Mouse tissue physiology: vascular & interstitial volume fractions (Mandikian et al., AAPS 2018)
- Two-pore model; residual blood fraction per tissue; FcRn K_D

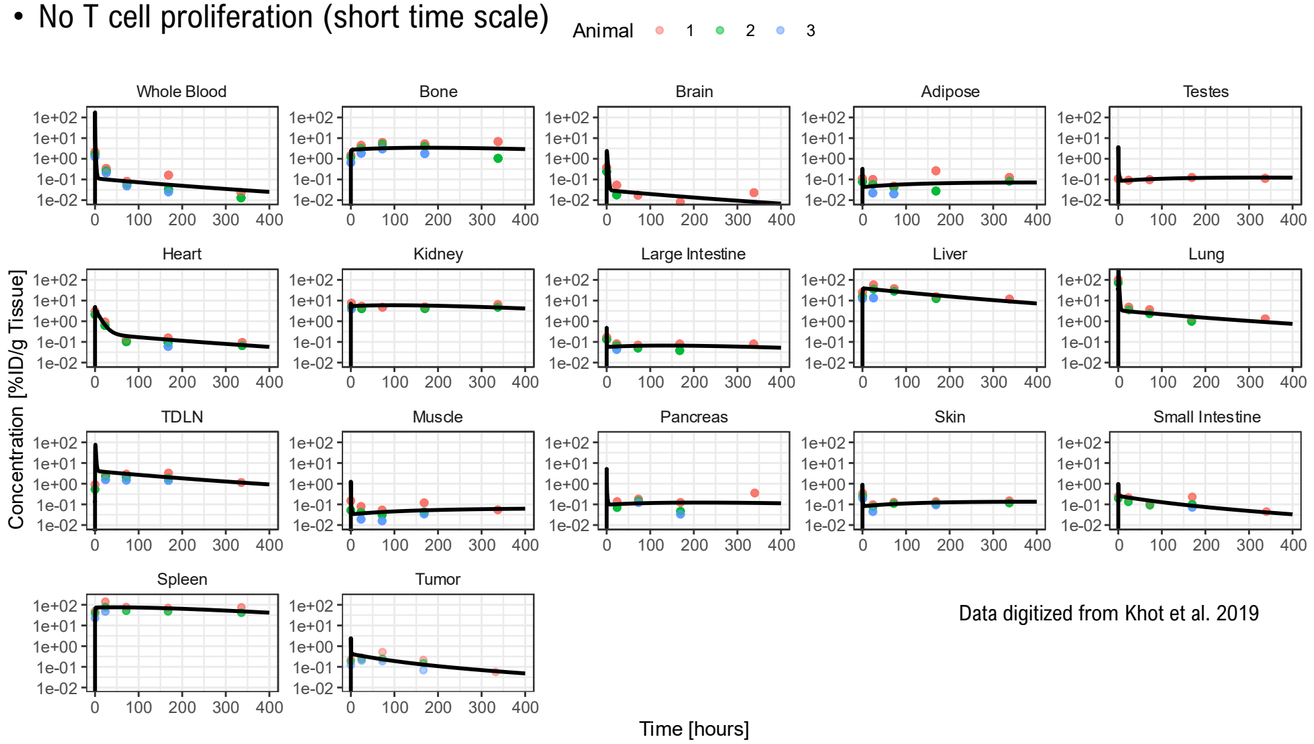
4. Capturing the distribution of gD/CD3 antibody
- CD3 receptor turnover
- Migration rate ratio for active to resting T cell

5. Capturing the distribution of HER2/CD3
- HER2 turnover and expression



Two organ-specific parameters to calibrate:
- T cell transmigration rate: T cell migration from vascular to extravascular space
- T cell retention factor: accounting for exogenous T cell retention in the organ

Major limitations / assumptions:
- T cell elimination only in the lung vascular space
- No T cell proliferation (short time scale)



Data digitized from Khot et al. 2019

The model was able to capture the distribution of exogenously administered T cells, which allows the base understanding of T cell distribution.

Building a bsTCE model — the typical workflow

TCE models share a common framework of **reusable mechanistic building blocks**; complexity grows on two axes — the workflow evolving from in vitro to human, and the granularity each question demands.

Describe the biology (MoA)

→ *which mechanisms to include?*

- *dimers formation*
- *trimer formation*
- *internalisation and degradation*
- *cytokine release*
- *target-cell killing*
- *T-cell activation*

Calibrate *in vitro* assays

→ *calibration is a multi scenario problem*

- n concentrations
- m target cell densities
- o E:T ratios
- p time points
- q cell lines / receptor densities
- r T-cell sources
- s cytokines

Model compartmentalization

→ *what spatial resolution?*

- *Compartmental PK*
- *mPBPK*
- *WB PBPK*

Transfer model to animal

→ *what stays mechanistic?*

- preserve mechanistic core/MoA relationships from *in vitro*

→ *what becomes species-specific?*

- PK / TMDD physiology
- Cell and receptor abundances
- *Cell dynamics and trafficking*
- *Cytokine network*
- *Immunogenicity (ADA)*

→ *re-calibrate considering species physiology*

- n doses
- m routes
- o time points
- p compartments
- q observables

Translate to human

→ *what changes across species?*

- Same mechanisms, but with human parameterization

→ *analyze and predict*

- Doses and schedule
- Efficacy/response
- Safety (CRS, cytopenias)
- Target engagement
- Biomarkers
- Patient variability
- Special populations
- Therapeutic window



3. One solution for multiple purpose

Subtitle

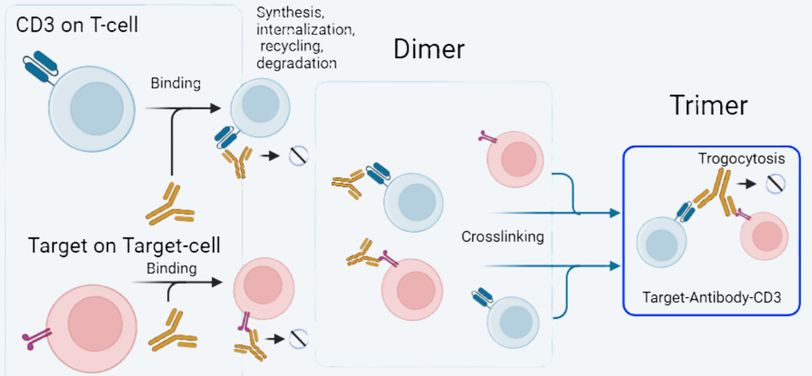
STEP 1 — In vitro calibration, a critical step

MoBi Modules

- 01 - Extension to binding T1
- 01 - Extension to binding TcellT1
- 01 - Extension to trimer binding T1 - TcellT1
- 02 - Internalization-DRUG-Target1
- 02 - Internalization-DRUG-TcellTarget1
- 03 - TcellActivation
- 04 - TargetCellKilling
- 05 - Tcell derived cytokine production
- DRUG
- InVitro

Model

- *dimers formation*
- *trimer formation*
- *internalisation and degradation*
- *cytokine release*
- *target-cell killing*
- *T-cell activation*



The multi scenario problem

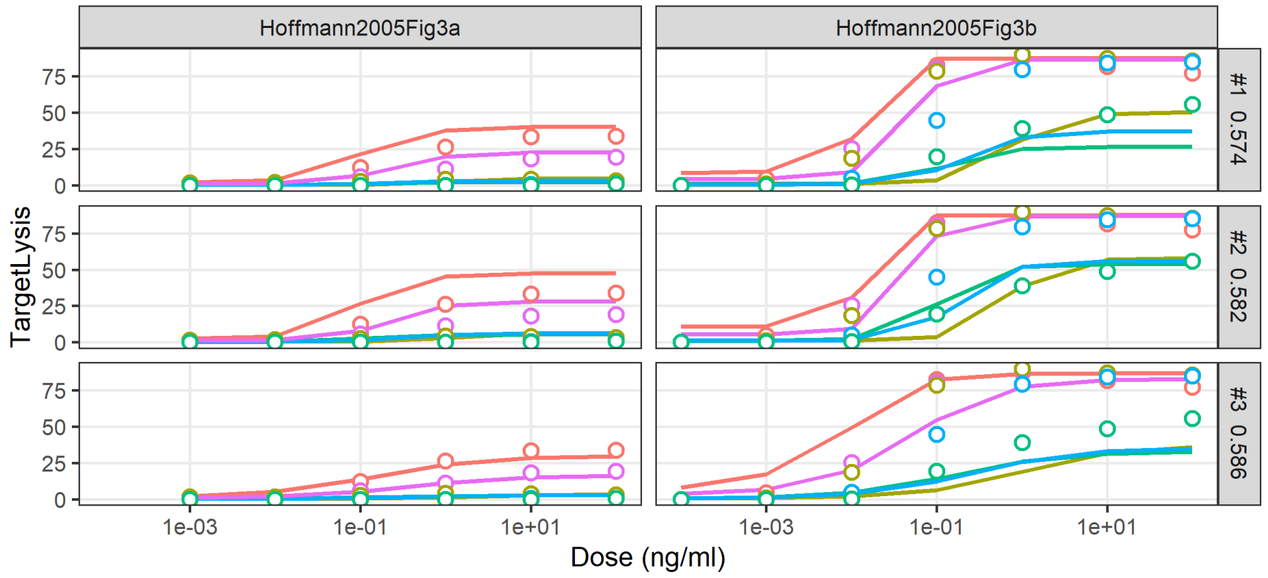
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- s cytokines

Pipeline

- Scenario definitions and calibration data
- Precalibration, exploration LHC sampling
- Parameter identification
- Practical identifiability

BlinaInVitroETratio - TargetLysis: the 3 best sets against the data

Lines = simulated, hollow points = observed
Rows = the best sets (rank, normalised RMSE)



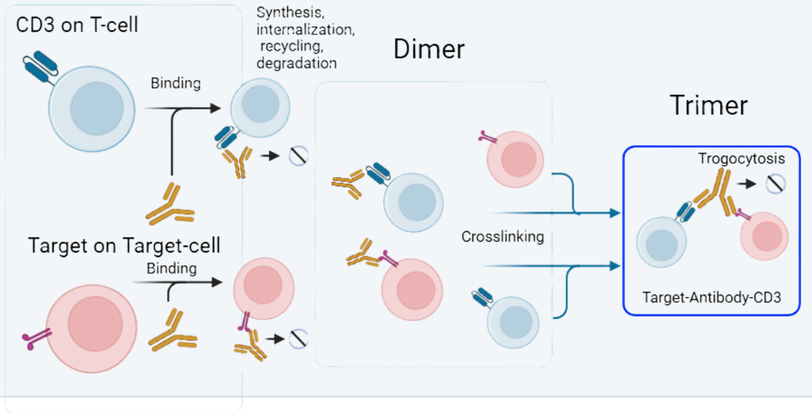
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The multi scenario problem

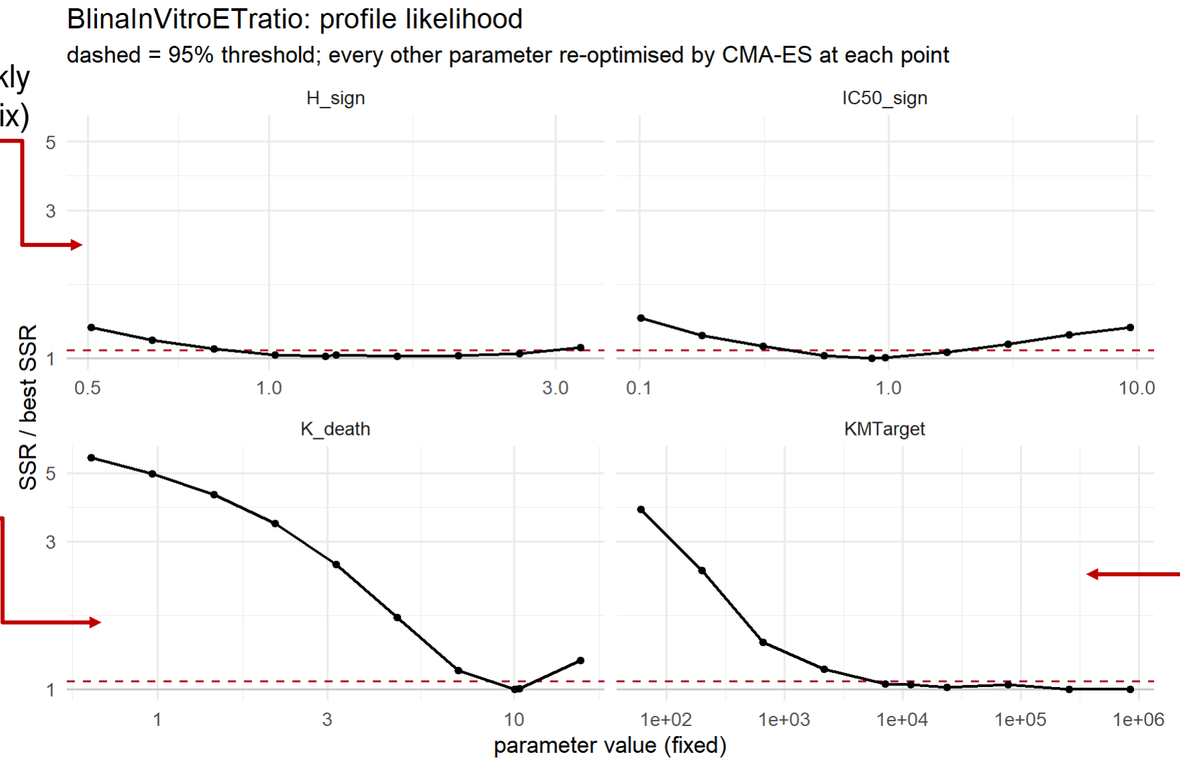
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Pipeline

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- Practical identifiability

Example of weakly identifiable parameter (->fix)

Example of a practically identifiable parameter



Example of a practically non-identifiable parameter (-> simplify)

STEP 2 — Compartmentalization defines the modeling sweet spot

Each spatial resolution trades physiological detail against data economy. The right one depends on the question addressed — which is why the **biology/MoA must be portable across all them.**

In vitro setting

Scope
- Single well-mixed volume;
- Drug concentration is fixed input
- MoA as relevant for each assay

Best for
- Calibration binding network
- Prototyping uncertain mechanisms
- Calibration against in vitro data package

Limitations
- Only reduced system (not all mechanisms are included) compared to in vivo
- Needs combination of models and scenarios

Compartmental PK

Best for
- Integration with pre-existing popPK model
- NLMEM modeling of PK/PD/ER

Limitations
- Empirical, reductionist, limited extrapolation

Minimal PBPK

Best for
- Middle-out modeling: key mechanisms but data driven
- Fast enough for multidimensional PI, LHS, sensitivity

Limitations
- Tissues are lumped (not resolved by organ)
- Local phenomena (target depletion) can be masked by lumping

Whole-body PBPK

Best for:
- Extrapolation by mechanism within an established platform
- Mechanistic translation: swap the physiology, keep the drug parameters
- “PBPK questions”: DDI, special populations, route and formulation changes

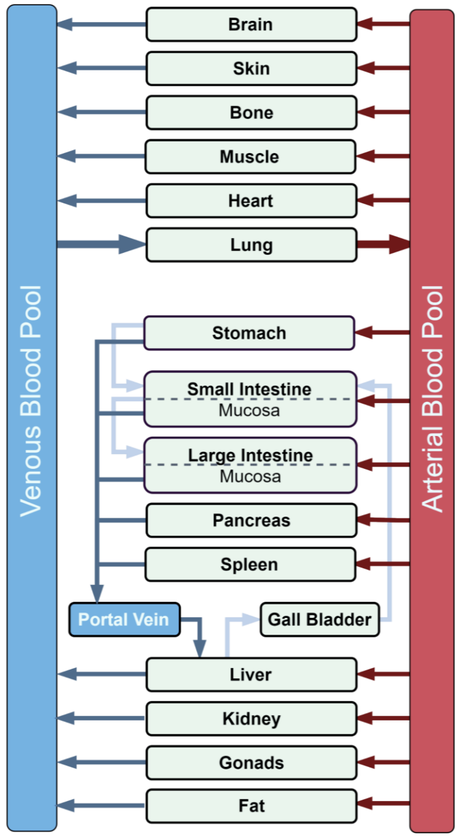
Limitations
- Complex models → High computational cost
- Processes and parameters are fixed from literature; limited flexibility
- Over-parameterisation = unidentifiability

STEP 3 — Transfer to animal, include physiology

A **modular approach guarantees identical structure across contexts.** Not four similar models: one model, different compartmentalization and/or species.

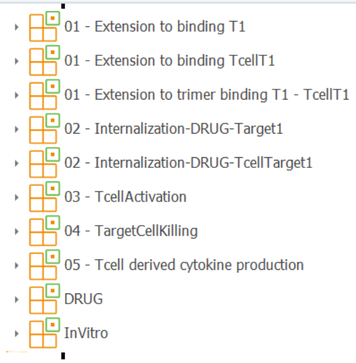
Define spatial resolution: compartmental PK, mPBPK, WB PBPK

Generic whole-body PBPK platform in PKSim and extendable in MoBi

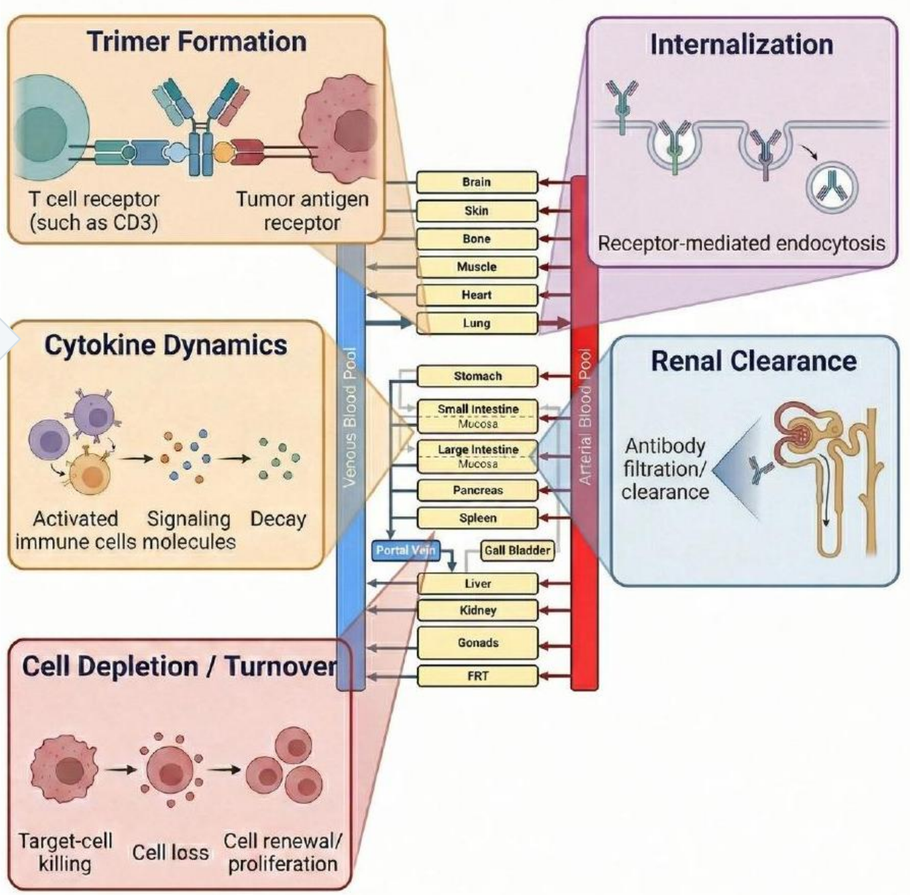


Niederalt et al. J Pharmacokinet Pharmacodyn (2018) 45: 235.

The **same MoBi modules** — amount-based, spatially agnostic — **are instantiated in every (plausible) container.**



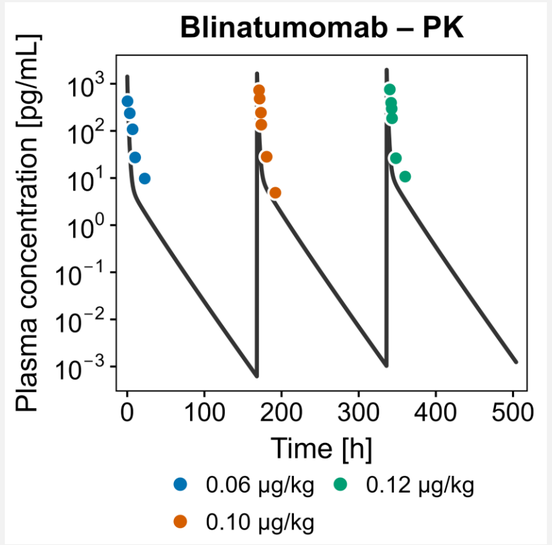
Reuse the same model and refit by species: mouse or NHP

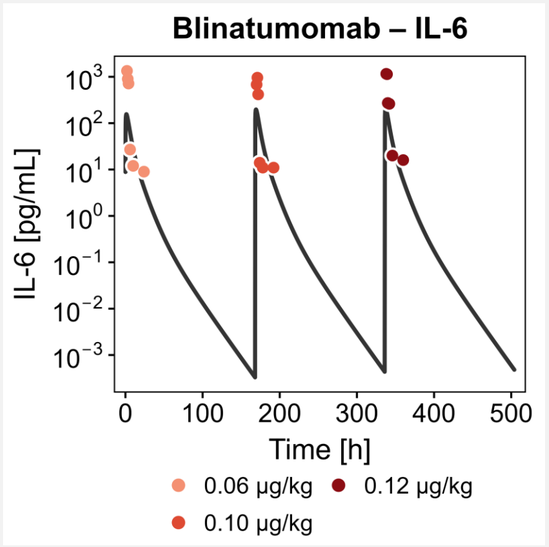


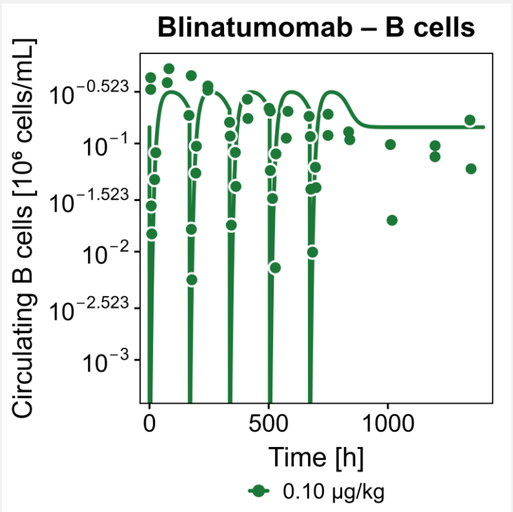
STEP 3 to 4 — Translate Monkey to Human

Practical Applicability: Blinatumomab

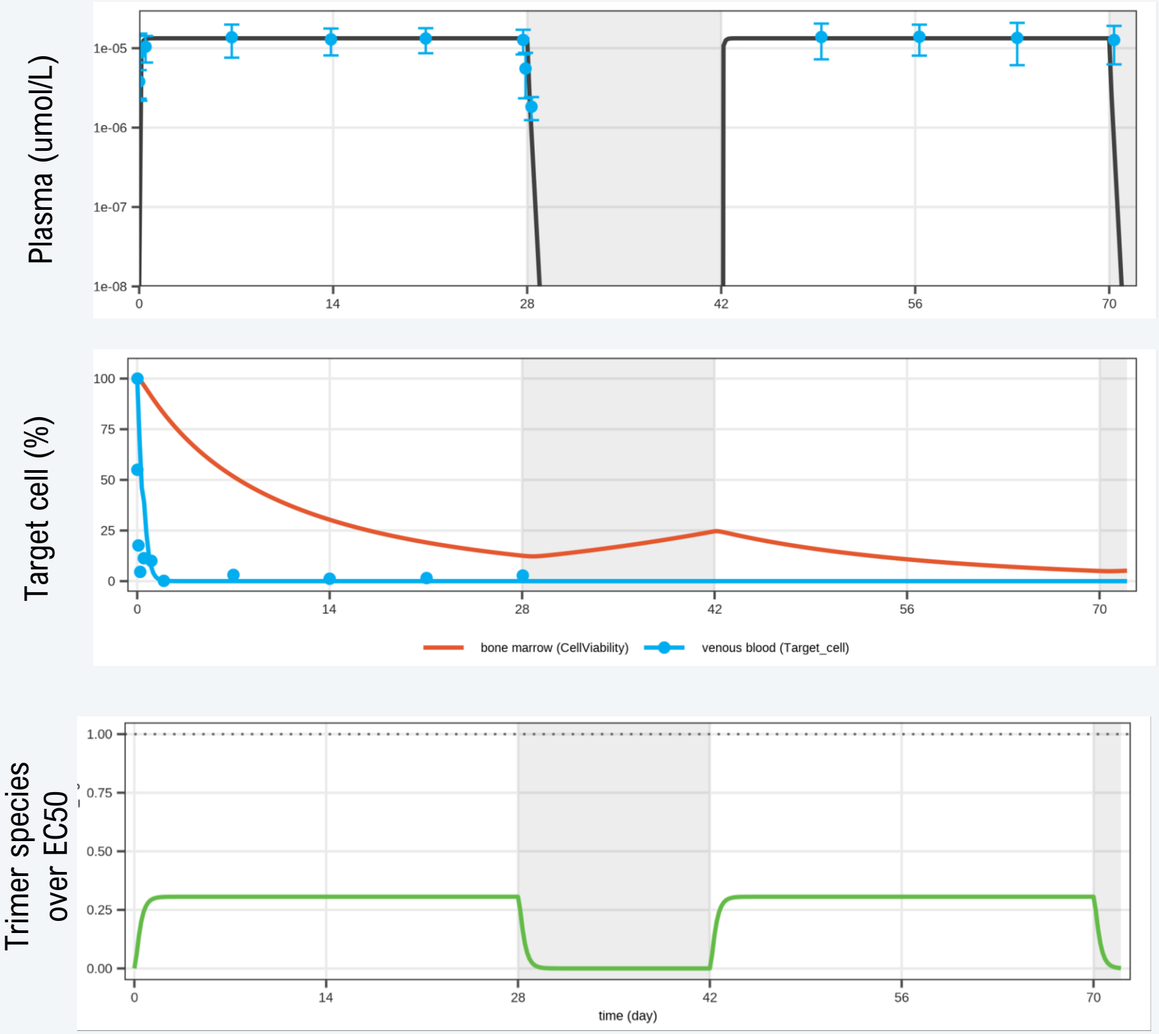
Monkey







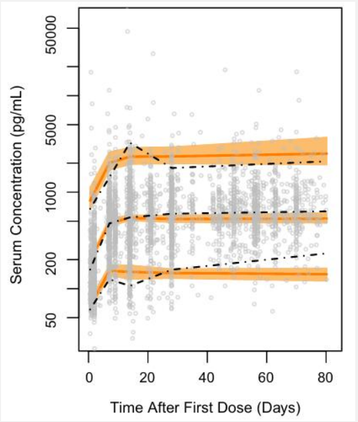
Human



STEP 4 —Human, explore and predict

Clinical Applicability: Blinatumomab (Human) – other scenarios/models

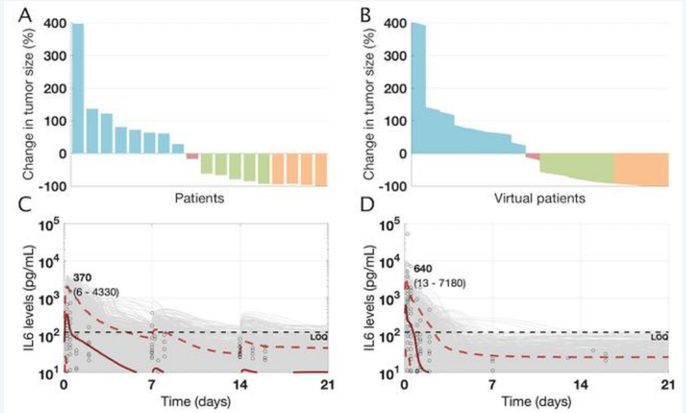
1 Comp model: Reuse parameters of popPK model - Clements et al (2020)



Parameter	Final model (mean (RSE, % [95% CI])
Pharmacokinetic parameters	
Volume (L)	5.98 (8.86) [5.14–6.98]
CL (L/h)	2.22 (2.95) [2.08–2.35]
Effect of BSA on CL (θ)[a]	0.620 (12.7) [0.46–0.76]
Inter-individual variability (%CV)	
ωCL	47.6 (16.1) [38.1–54.2]
ωEPS	64.3 (14.5) [55.0–73.0]
Residual variability (%CV)	55.9 (3.99) [52.6–61.5]

mPBPK/QSP model - Hosseini et al. (2020)

B-cells and T-cells are represented in the PB, lymphoid tissues (bone marrow—BM, lymph nodes—LN, and spleen), and when appropriate, a lymph-node NHL tumor.



- mPBPK – fitting translation
- Fitting PK, PD response
- Virtual twins

PBPK model: Assess of cytokine release CYP – DDI - Xu et al (2015)

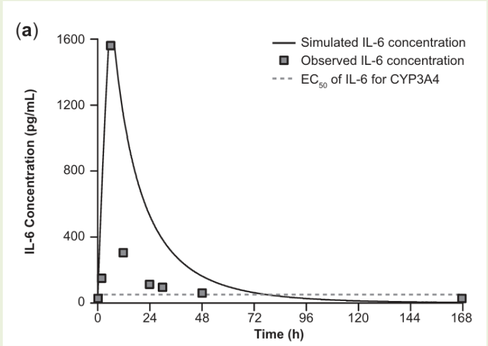


Table 3 Predicted maximum IL-6–mediated drug interaction on substrates of CYP3A4/1A2/2C9 based on the mean IL-6 profile[a]

Substrate	CYP450 affected	Mean AUC ratio (95% CI)	Mean C_{max} ratio (95% CI)
Simvastatin	3A4	1.9 (1.8–2.0)	1.7 (1.6–1.8)
Midazolam	3A4	1.7 (1.6–1.8)	1.2 (1.1–1.3)
Theophylline	1A2	1.1 (1.0–1.1)	1.0 (1.0–1.0)
Caffeine	1A2	1.2 (1.1–1.3)	1.0 (1.0–1.1)
(S)-Warfarin	2C9	1.2 (1.0–1.4)	1.0 (1.0–1.0)

OSP as PBPK-QSP Platform — an integrated open-source ecosystem

The OSP platform's modular architecture enables model reuse across contexts, extension or modification to fit each scientific question, and a range of analyses via an R-based MIDD ecosystem.

Pharmaverse:
Clinical data handling
and reporting

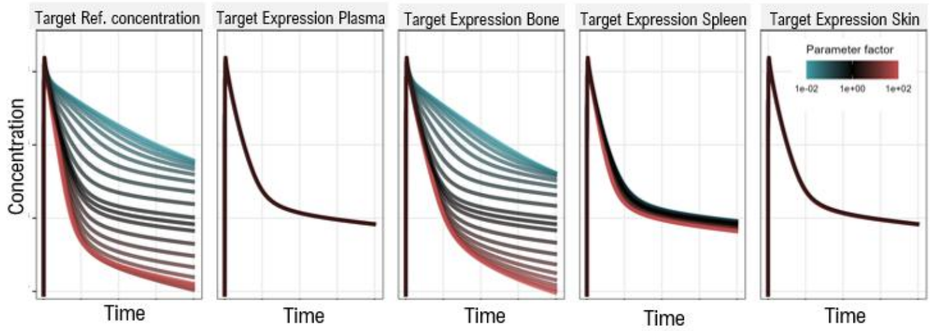


PBPK Modeling

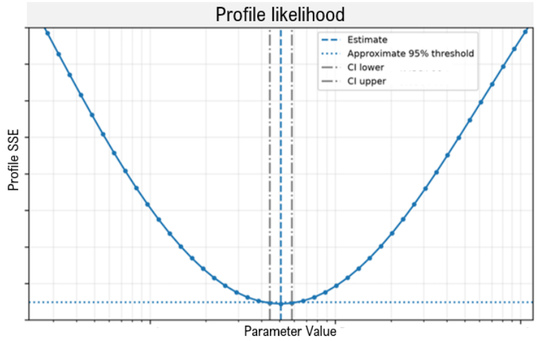
Empirical modeling (PK, popPK, PKPD ER)

QSP modeling

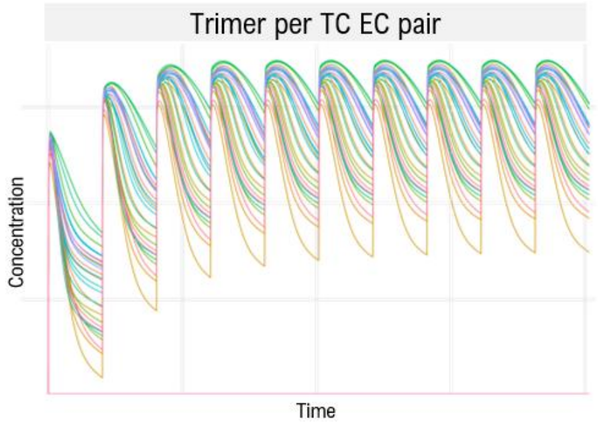
Sensitivity Analysis

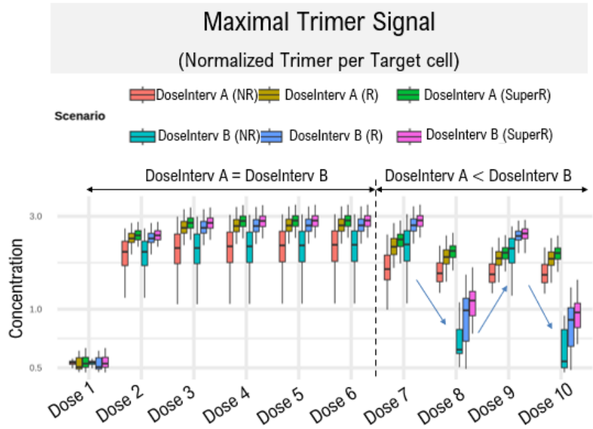


Uncertainty/Identifiability Analysis



Virtual Twins/ Virtual Populations







4. Conclusions & Next Steps

Subtitle

Next steps

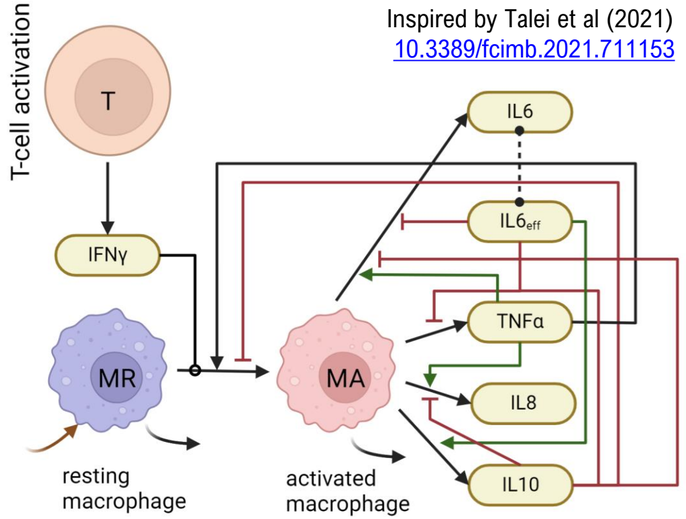
Prototyped in MoBi and documented, not yet calibrated into the platform

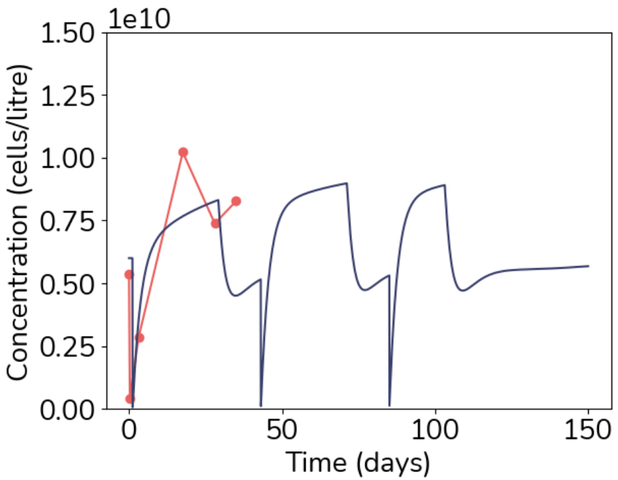
Multi state T-cell activation

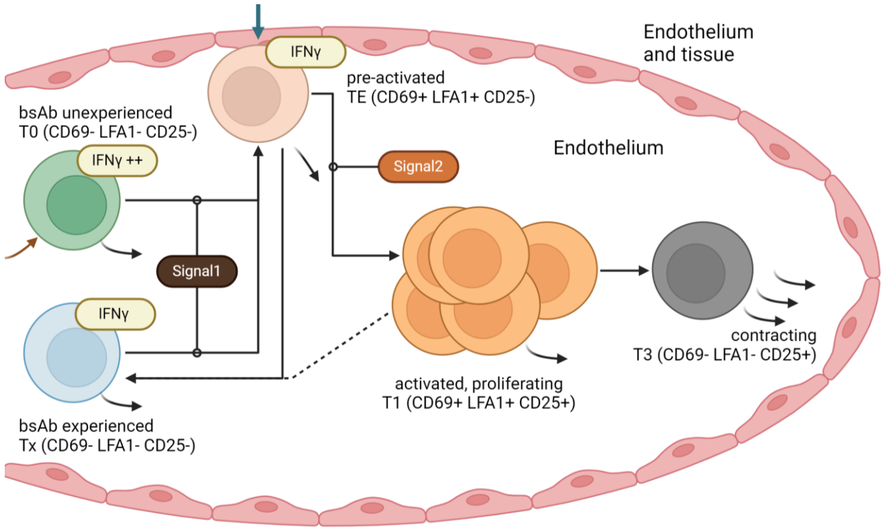
Five states: naïve, engaged, cytotoxic T2, terminally activated T3, exhausted Tx. The trimer signal drives the transitions and T2 drives the killing, so the effector-to-target ratio stops being an input and becomes a state — which is what re-dosing and treatment-free intervals need.

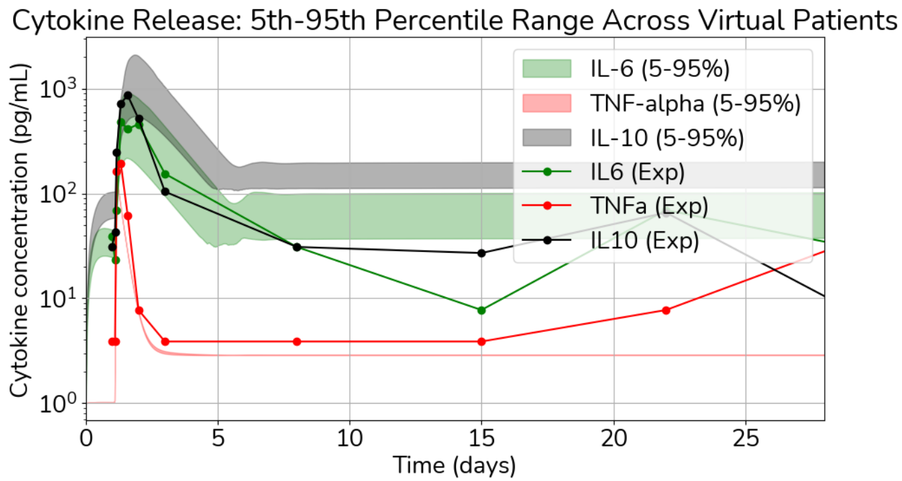
Detailed cytokine network

IFN-γ, IL-2 and TNF-α on one shared signal-driven ODE. These are the readouts behind cytokine release risk, the MABEL argument and the rationale for step dosing. This comprises other effectors.









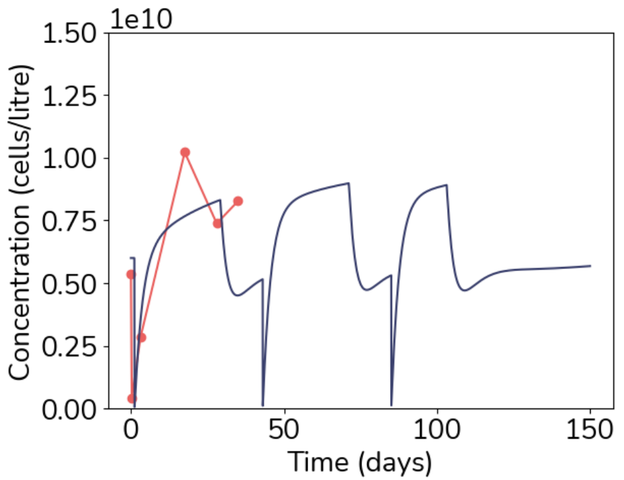
Also queued: cross-species scaling (cyno → human, on the structure that already holds the physiology) and MoBi.R, so a plugin is assembled from a script rather than by hand.

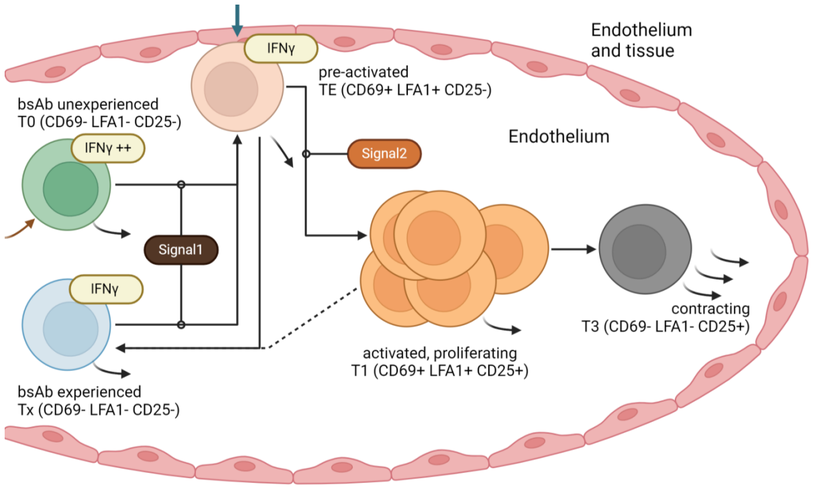
Next steps

Prototyped in MoBi and documented, not yet calibrated into the platform

Multi state T-cell activation

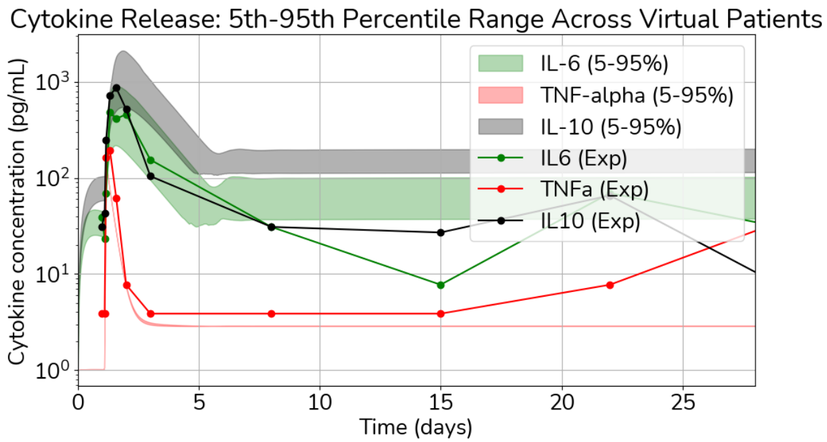
Five states: naïve, engaged, cytotoxic T2, terminally activated T3, exhausted Tx. The trimer signal drives the transitions and T2 drives the killing, so the effector-to-target ratio stops being an input and becomes a state — which is what re-dosing and treatment-free intervals need.



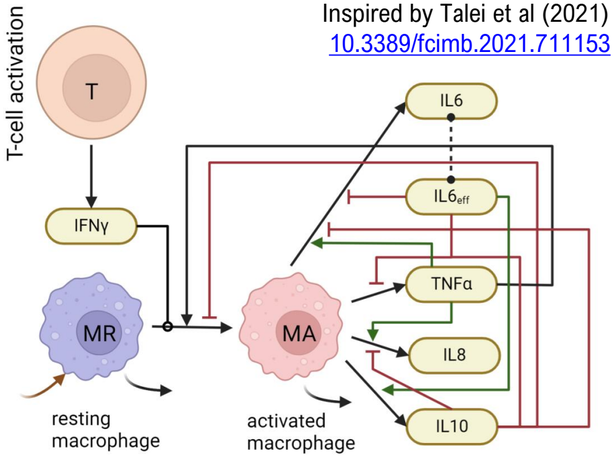


Detailed cytokine network

IFN-γ, IL-2 and TNF-α on one shared signal-driven ODE. These are the readouts behind cytokine release risk, the MABEL argument and the rationale for step dosing. This comprises other effectors.



Inspired by Talei et al (2021)
10.3389/fcimb.2021.711153



Also queued: cross-species scaling (cyno → human, on the structure that already holds the physiology) and MoBi.R, so a plugin is assembled from a script rather than by hand.

Conclusions

1. **A parameter keeps its definition across spatial structures**
   because the modules are written in concentration terms and know nothing about the container they sit in. Only the volume differs.

2. **The heavy lifting is done once**
   in vitro, across 65 scenarios at once — and the estimates are then used unchanged in the one-compartment, mPBPK and whole-body models.

3. **The fitting is a specification, not a script**
   declared in Excel and executed by a pipeline that contains nothing about this project. Adapting it to another molecule, assay or structure is editing rows.

4. **Whole-body PBPK enables mechanism-based predictions and extrapolation**
   It enables readout of latent variables, and exposes physiological mechanisms for perturbation that more empirical models hide.

5. **For the next modality, only the extension modules and specifications change**
   which is what makes this a platform rather than a single-use model.

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THANK YOU



Photo: Grote Markt, Antwerp, Belgium

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