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Physiologically-Based Pharmacokinetics (PBPK) for the Modelling of Bispecific Monoclonal Antibody Tumour Targeting Specificity

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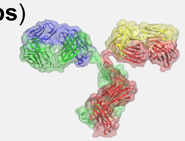
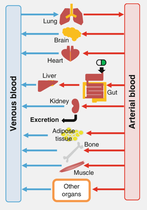
PURPOSE

- The FDA has directed replacing the animal testing of biologics with *in silico* modelling and simulation[1]
- Bispecific mAbs (**bsmAbs**) are expected to improve the tumour-targeting specificity of antibody-drug conjugates (ADCs)



- While the bsmAbs can be highly specific for the cells that co-express two different antigens, their tissue targeting specificity *in vivo* is largely unknown.

OBJECTIVE



- We use Physiologically-based pharmacokinetics (**PBPK**) to evaluate the likely *in vivo* tumour-targeting specificity of bsmAbs
- We focus on the impact that the bsmAb dose, affinities, and target concentrations are likely to have on the tumour targeting specificity and healthy tissue exposure of this modality.

METHODS

- **EGFR** and **HER2** served as model antigens for the bsmAb modelling. Their tissue concentrations were derived from protein mass spectrometric data[2]
- The bsmAb properties were based on cetuximab and trastuzumab, and the cross-linking reaction followed the 2D-kinetics[3].
- The pharmacokinetic model was run in Matlab SimBiology, and it was built using computer-assisted PBPK model assembly tools[4].

RESULTS

All organs of the body, including the solid tumour, were modelled to have 1:1:1:1 of cells expressing just one of the targets, both or none at all, as outlined in Figure 1.

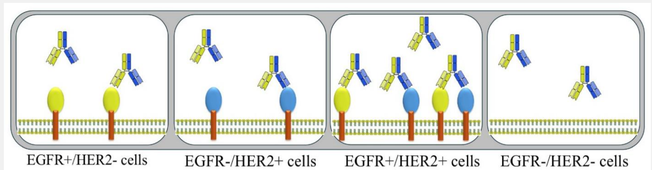


Figure 1. EGFR and HER2- expressing cells' interactions with the bsmAb

The predicted target occupancy time courses in the solid tumour for an isotype control, two parent mAbs and the bsmAb, at 0.1, 1, 10 mg/kg single IV doses, are shown in Figure 2

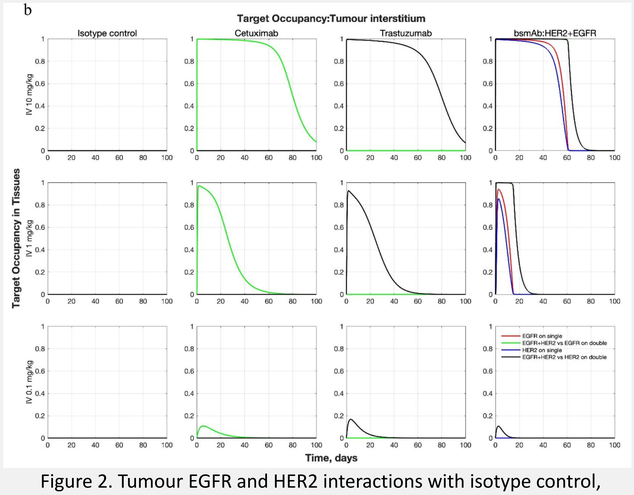


Figure 2. Tumour EGFR and HER2 interactions with isotype control, monospecific parent mAbs, and the bsmAb

- bsmAb is likely to afford specific engagement dual-positive cells in the tumour, *but this only happens at very low drug concentrations, when monovalent interactions no longer form*
- At the same time, relatively high bsmAb doses ≥1 mg/kg are needed to achieve sufficient level of target engagement in the solid tumour.

The relative uptake of mono- and bispecific mAbs in selected tissues and cell types is shown in Figure 3. This combines both non-specific and target-mediated uptake.

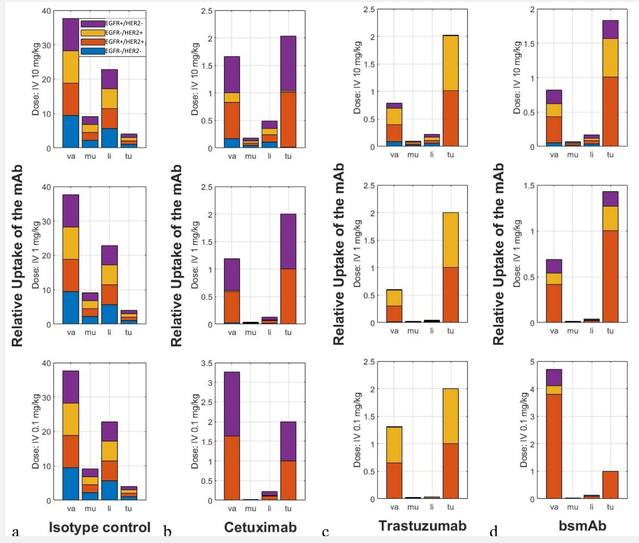


Figure 3. The relative uptake of isotype control, monospecific parent mAbs, and the bsmAb in the four different cell types in the vascular endothelium, skeletal muscle, liver and solid tumour at 0.1, 1 and 10 mg/kg IV dose. va-vasculature, mu-skeletal muscle, li-liver, tu-tumour.

At each dose, the amount of bsmAb catabolised by dual-positive tumour cells is defined as the benchmark.

- The isotype control mAb is taken up in equal amounts by all cell types in a tissue, regardless of antigen expression status.
- Monospecific mAbs are predominantly taken up by cells expressing the respective antigens, but there is no discrimination between single- and dual-positive cells.
- Dual-positive tumour cell targeting is only observed at the lowest dose, when the drug concentration is very low
- Depending on the dose, about 0.1-1 % of the bsmAb ends up in the tumour, suggesting that the therapeutic window remains narrow.

CONCLUSIONS

Exquisite dual-positive cell-binding specificity of bsmAbs *in vitro* is unlikely to translate into equalluy exquisite tumour tissue targeting specificity *in vivo*.

PBPK modelling confirms that only a very small fraction of the dosed mAb (mono- or bispecific) is likely to reach solid tumours.

- Whilst this can be negligible in the case of unconjugated bsmAbs, in the case of ADCs, this can result in dose-limiting toxicity.
- More rapid mAb absorption and uptake in healthy tissues by non-specific and target-mediated routes is causing the imbalance
- Avidity-enhanced in vitro binding of bsmAbs to the cells is unlikely to translate to similarly enhanced efficacy in vivo, but modelling can help to optimise the drug properties
- Quantitative mechanistic modelling can support and guide the development of new modalities from bench to bedside. This includes target evaluation and optimisation of drug affinities and dosing for optimal exposure[5].

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