Development of alternative in vitro and ex vivo models for testing of inhalable antibiotics – “InhalAb”

S. Wronski[1*], K. Schwarz[1*], D. Ritter[1], J. Knebel[1], K. Blümlein[1], N. Sonnenschein[1], S. Cleeves[1], L. Müller[1], N. Nowak[1], M. Niehof[1], K. Sewald[1], T. Hansen[1], A. Braun[1]

[1] Fraunhofer Institute for Toxicology and Experimental Medicine ITEM, Member of the German Center for Lung Research (DZL), Biomedical Research in Endstage and Obstructive Lung Disease (BREATH) research network, Member of Fraunhofer international Consortium for Anti-Infective Research (iCAIR), Hannover, Germany
* contributed equally

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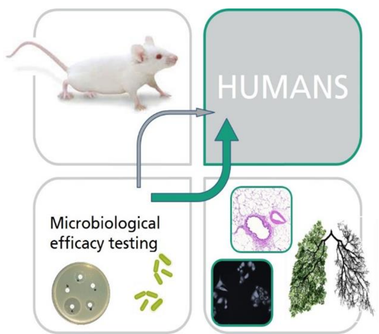
InhalAB (03L0135)

Aim of the project

The project aim is to establish a test battery of in vitro and ex vivo lung models for preclinical testing of inhalable antibiotics as an alternative model to animal testing, which is associated with a high burden for the laboratory animals. This coordinated test battery shall be developed as a replacement for preclinical efficacy and toxicity studies as well as pharmacokinetic investigations of new inhalable antibiotics and formulations, aiming at optimization of drug targeting and dose-finding studies.



Toxicity testing
Efficacy testing
Pharmacokinetics

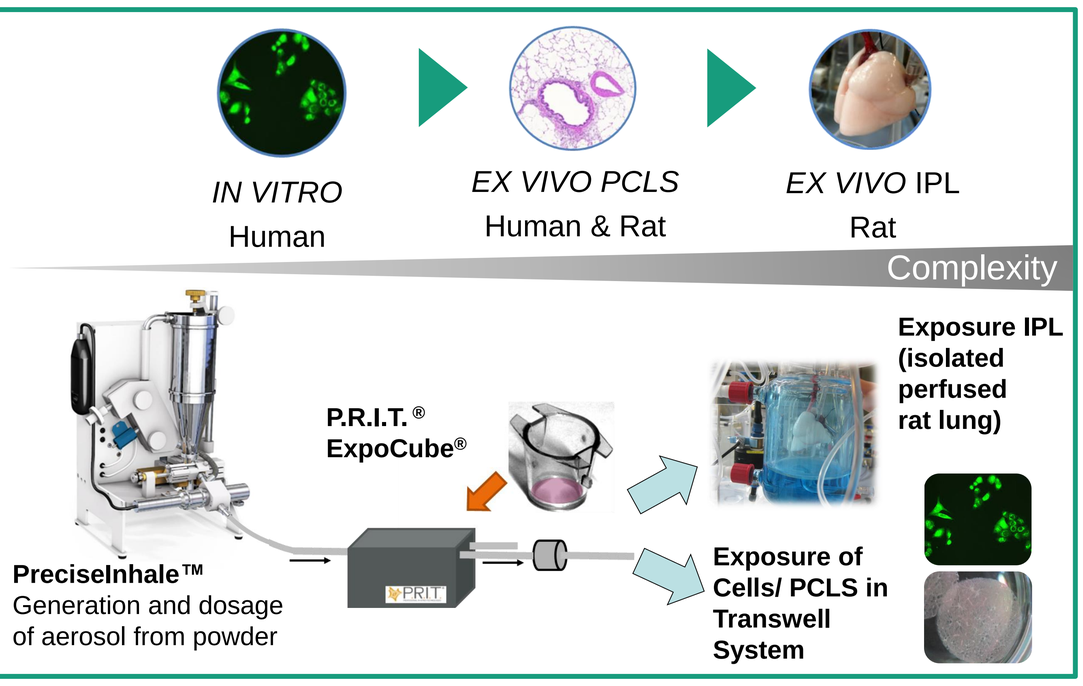
Therapeutic window & PBPK modelling

Methods

A test battery of alternative models with human cell lines, precision cut lung slices (PCLS) and isolated perfused rat lung (IPL) has been established for animal-free testing of toxicity, efficacy and pharmacokinetic analysis of inhalable antibiotics.

Inhalation application was established using air-liquid interface exposure of cells and PCLS via the P.R.I.T.® ExpoCube® using the reference antibiotics tobramycin and ciprofloxacin.

Efficacy of inhalation application was assessed in an ex vivo infection model in PCLS using *Pseudomonas aeruginosa* as a key lung pathogen. For PK analysis, human lung epithelial cells in vitro and IPLs ex vivo were exposed to inhalable antibiotic aerosols. Transfer rates and blood-tissue-distribution coefficient determined were used as input parameter for a first inhalation PBPK model (Physiological-based Pharmacokinetic model) recently developed by Fraunhofer ITEM.

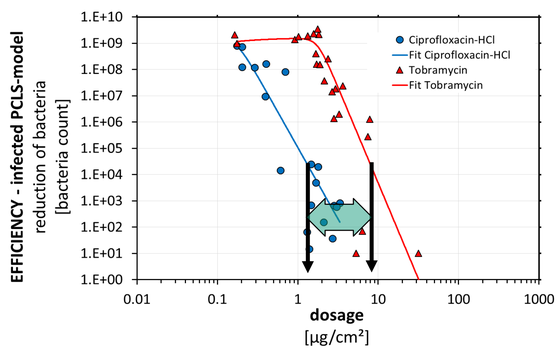


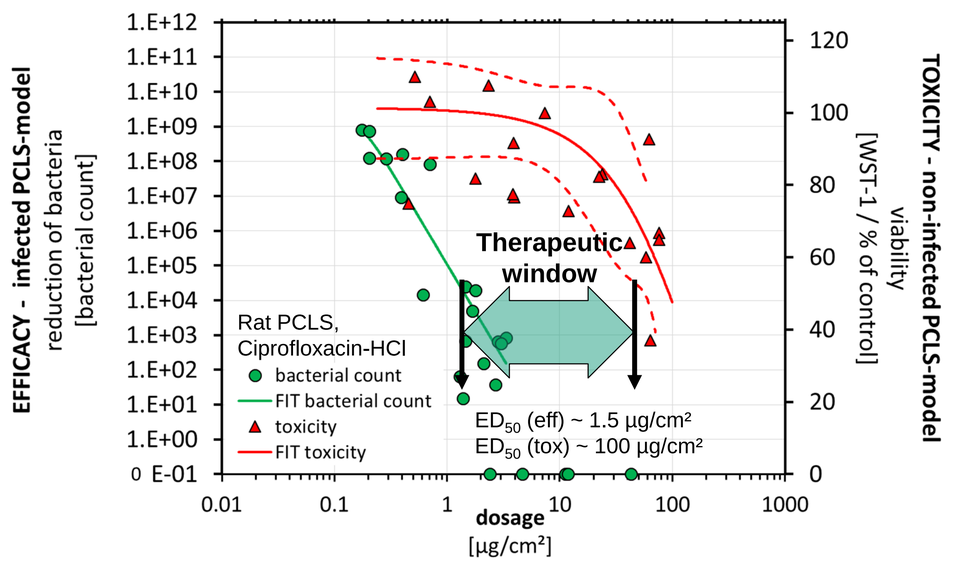
Results - *Local toxicity and efficacy testing*

In the PCLS model, comparative analysis of local toxicity and efficacy showed that a therapeutic window can be well represented: In the non-toxic range, both antibiotics led to an effective dose-dependent reduction of the bacterial load after infection with *Pseudomonas aeruginosa* and thus to the preservation of tissue viability.

Relative quantitative comparison of effective, mass/surface-based dosages values showed an about 5-fold higher value for tobramycin, which is in line with corresponding MIC–values.

A first rough approach to translate the efficacy data determined ex vivo into an effective human dose, showed promising results for both reference antibiotics.

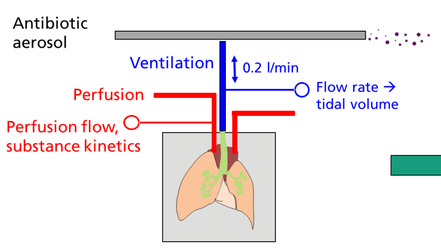




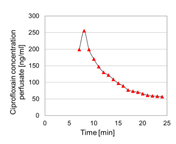
Results - *Pharmacokinetic analyses*

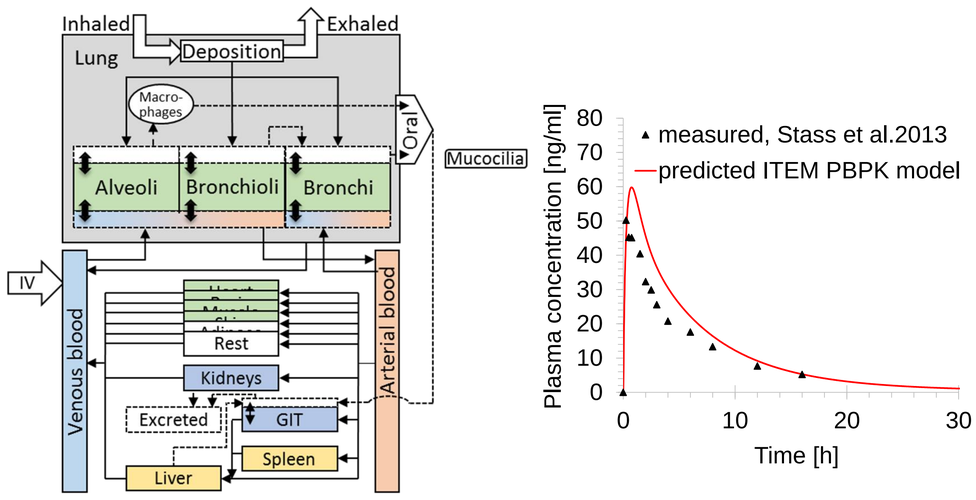
Besides determination of transfer rates, a first experimental design for analysis of the blood-tissue distribution in the IPL was developed, which, according to the current state of knowledge, allows for prediction of a human-relevant value.

Using these data as input parameter in the PBPK model, a good agreement of the predicted systemic concentration-time profile with the corresponding human data was observed for ciprofloxacin.



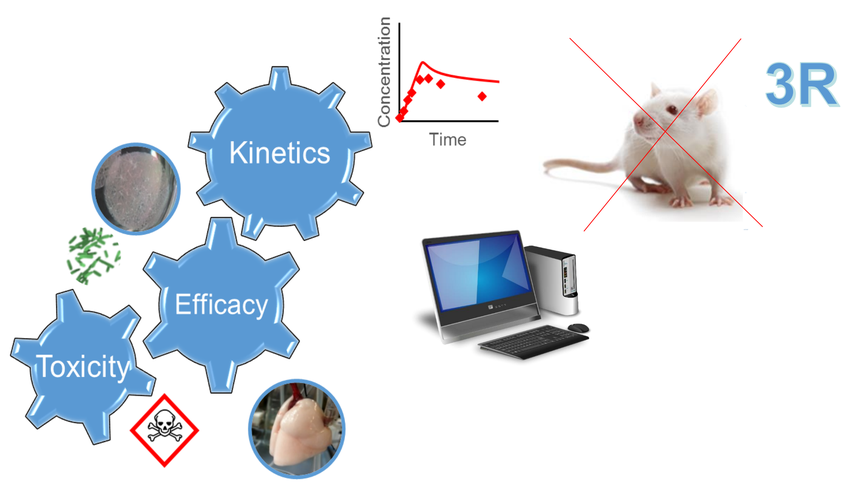
Transfer rate, blood-tissue-distribution





Summary and perspective

Alternative models of human cell lines, PCLS and IPL have been successfully established and allow for the determination of efficacy, toxicity and pharmacokinetic parameters for inhalable antibiotics. **Therefore, predictive preclinical data sets can be collected without animal testing in vivo.** The data collected here are an important step towards avoiding burdensome animal experiments for antibiotic testing.



Contact

sabine.wronski@item.fraunhofer.de, katharina.schwarz@item.fraunhofer.de