In vitro Pk/Pd modelling of antimycobacterial multi-drug regiments

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PhD candidate

HFS User Meeting
27/11/2019 Basel

Center for Infectious Diseases
Radboudumc

Radboudumc TB/NTM center of expertise

- ‘Dekkerswald’ sanatorium

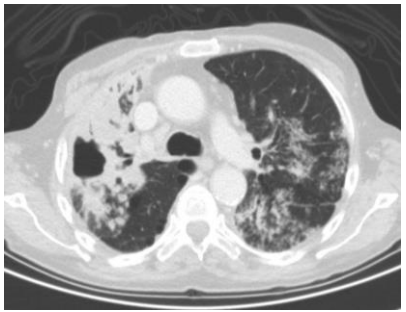
- Multidisciplinary team
  - Pulmonologists
  - Infectious Diseases
  - Pharmacists
  - Radiologists
  - Clinical microbiologists

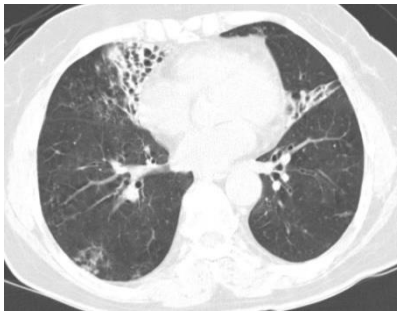
- Trial site & research laboratory

What?

NTM-PD – the 2019 situation

- Increasing prevalence
- No evidence-based treatment regimens
- Very low cure rates
  - *M. avium* complex: 50-70%
  - *M. abscessus*: 40-50%
- Recurrence rate: 40%





van Ingen J, et al. *Expert Rev Anti Infect Ther* 2013
Kwak N et al., *Eur Respir J* 2019
Zweijpfenning S, et al. Respir Med 2017

Treatment recommendations - MAC

American Thoracic Society

- *M. avium* complex
- **Rifampicin-Ethambutol-Azithromycin**

- *PLUS* 3 months of iv amikacin for severe (cavitary) disease

- With **surgical debulking** whenever feasible

Treatment duration

- Pulmonary disease: 12 months after culture conversion

Griffith DE, et al., *Am J Respir Crit Care Med* 2007
Floto RA, et al. *Thorax* 2016

Aim

Simulate Multi-antibiotic therapy with human PK

- Start at the **Rifampicin-Ethambutol-Azithromycin** combination
- Replace single drugs in this regimen

Intracellular culture in the HFS

- Use THP-1 cells as a model for human Macrophages
- Asesss intracellular and extracellular CFU counts

Assess resistance formation

- Sequence pathogen in the HFS
- Culture on antibiotic-containing plates

How?

PK/PD – the normal preclinical route

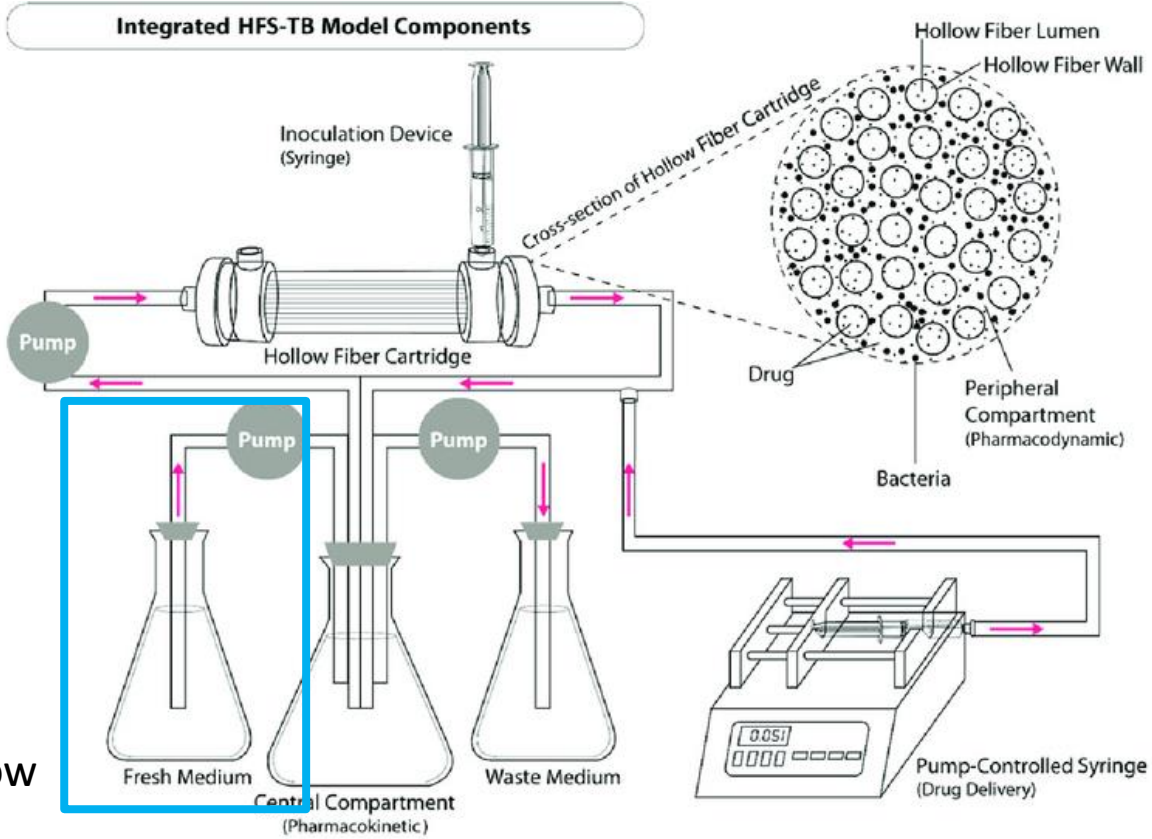
MIC
- Minimum inhibitory concentration / minimum bactericidal concentration
- **How active is this drug?**

SYN
- Checkerboard titrations / Fractional inhibitory concentration index
- **Is this drug synergystic with other antimycobacterial drugs?**

TKK
- Time-Kill kinetics assay
- **Is killing time/concentration/exposure dependent?**

MCF
- Antimycobacterial activity inside macrophages
- **Is this drug/regimen effective against intracellular mycobacteria?**

HFS
- Hollow fiber pharmacodynamic model / animal model
- **What is the optimal dose, dosing strategy and treatment duration?**

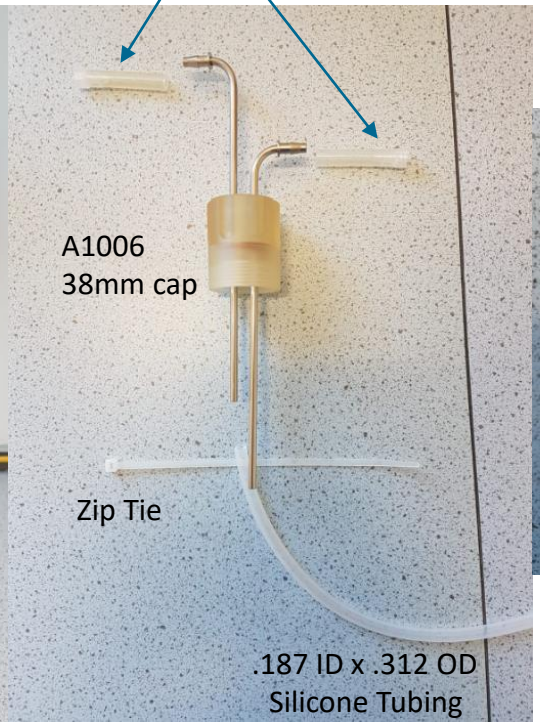

Integrated HFS-TB Model Components
Hollow Fiber Lumen
Hollow Fiber Wall
Inoculation Device
(Syringe)
Cross-section of Hollow Fiber Cartridge
Pump
Hollow Fiber Cartridge
Drug
Peripheral
Compartment
(Pharmacodynamic)
Bacteria
Pump
Pump
Fresh Medium
Central Compartment
(Pharmacokinetic)
Waste Medium
Pump-Controlled Syringe
(Drug Delivery)

Inflow

Inflow

4L Jug (autoclavable)

Plastic Luer connectors

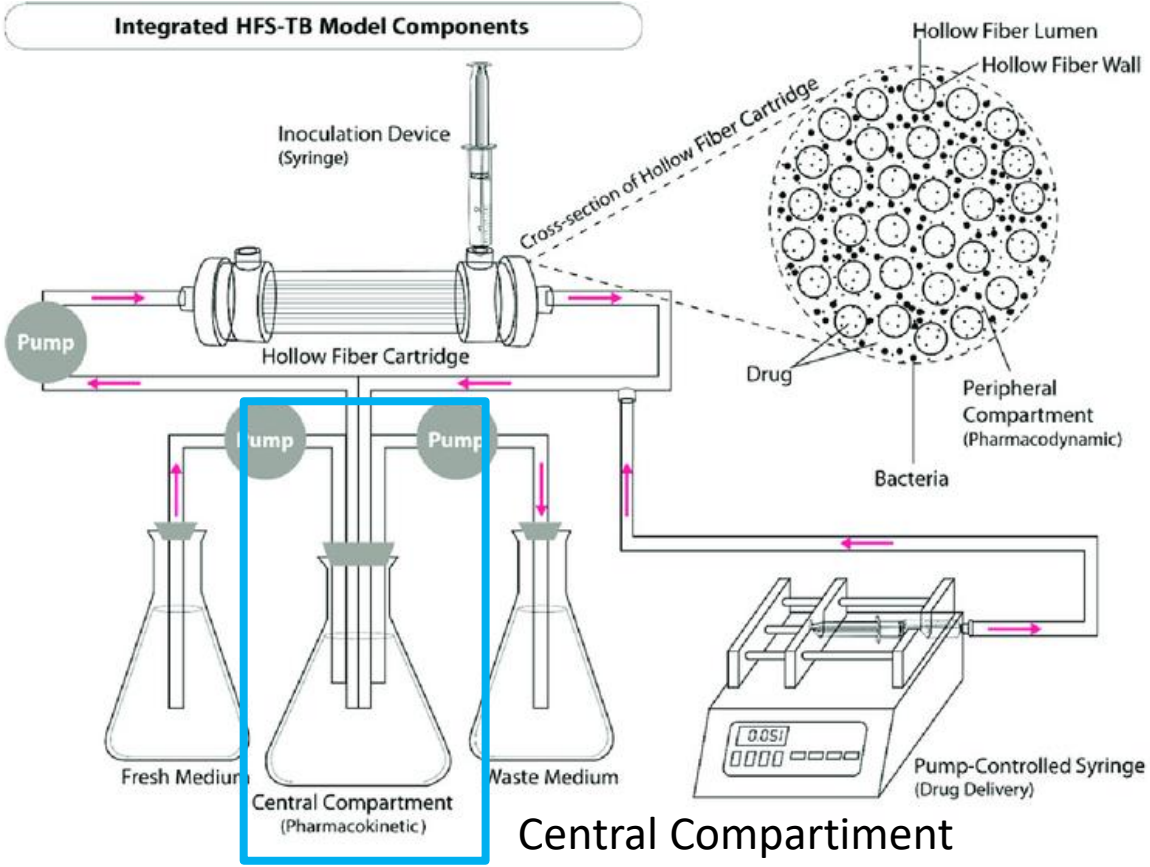


Culture Guard
0,2µm filter

Inflow

Watson Marlow peristaltic pumps in an N x 3 set up

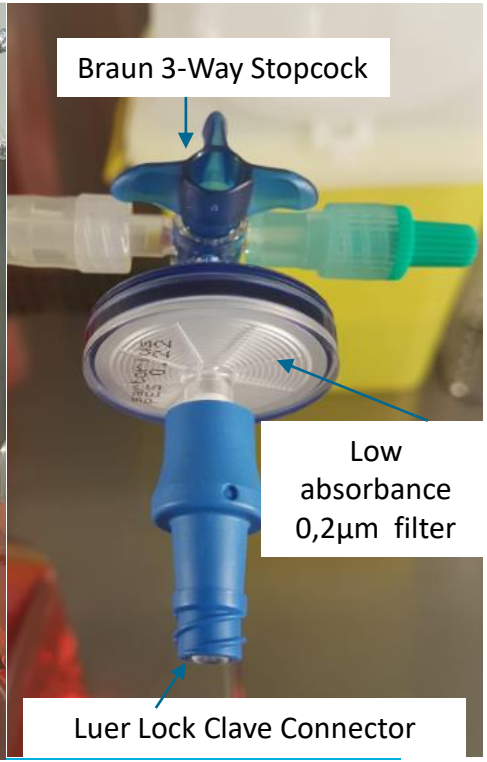
Watson Marlow Pumpsil cured silicone tubing 1.6mm ID



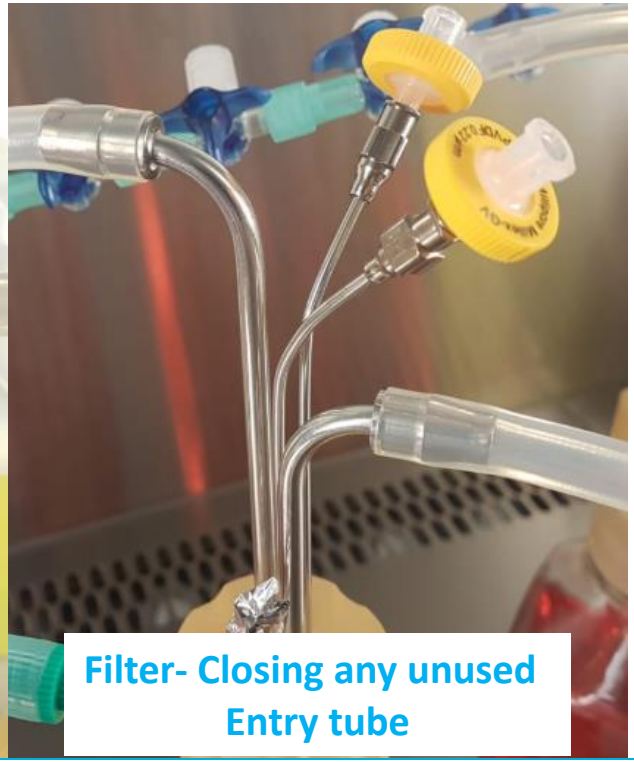
Central Compartiment

Central Compartiment

PK sampling Port

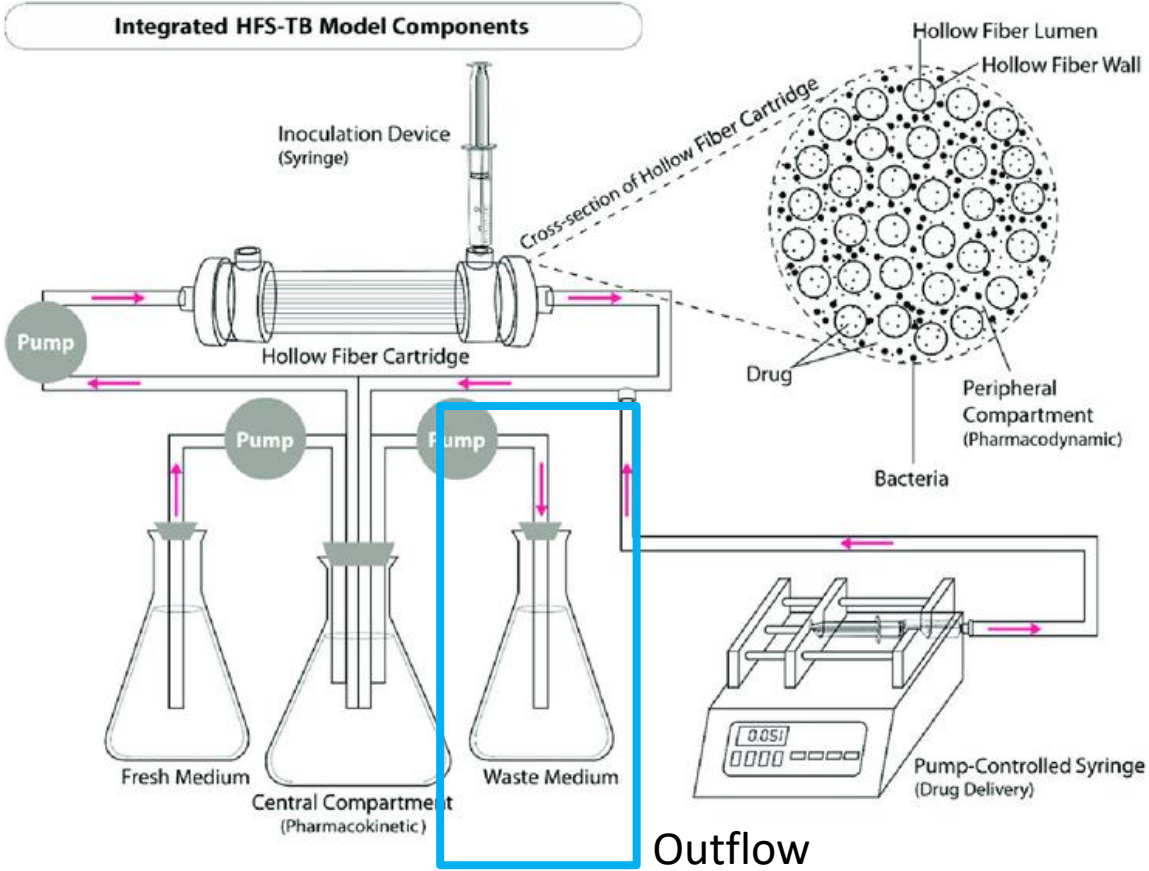


Central Compartiment



Central Compartiment

Full Setup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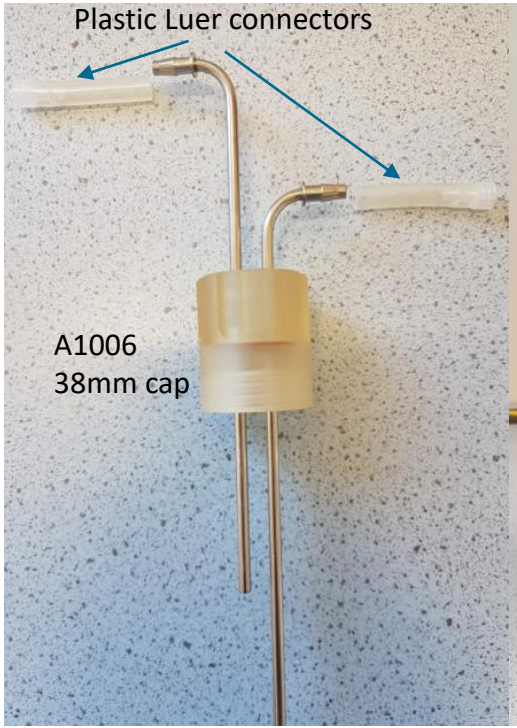
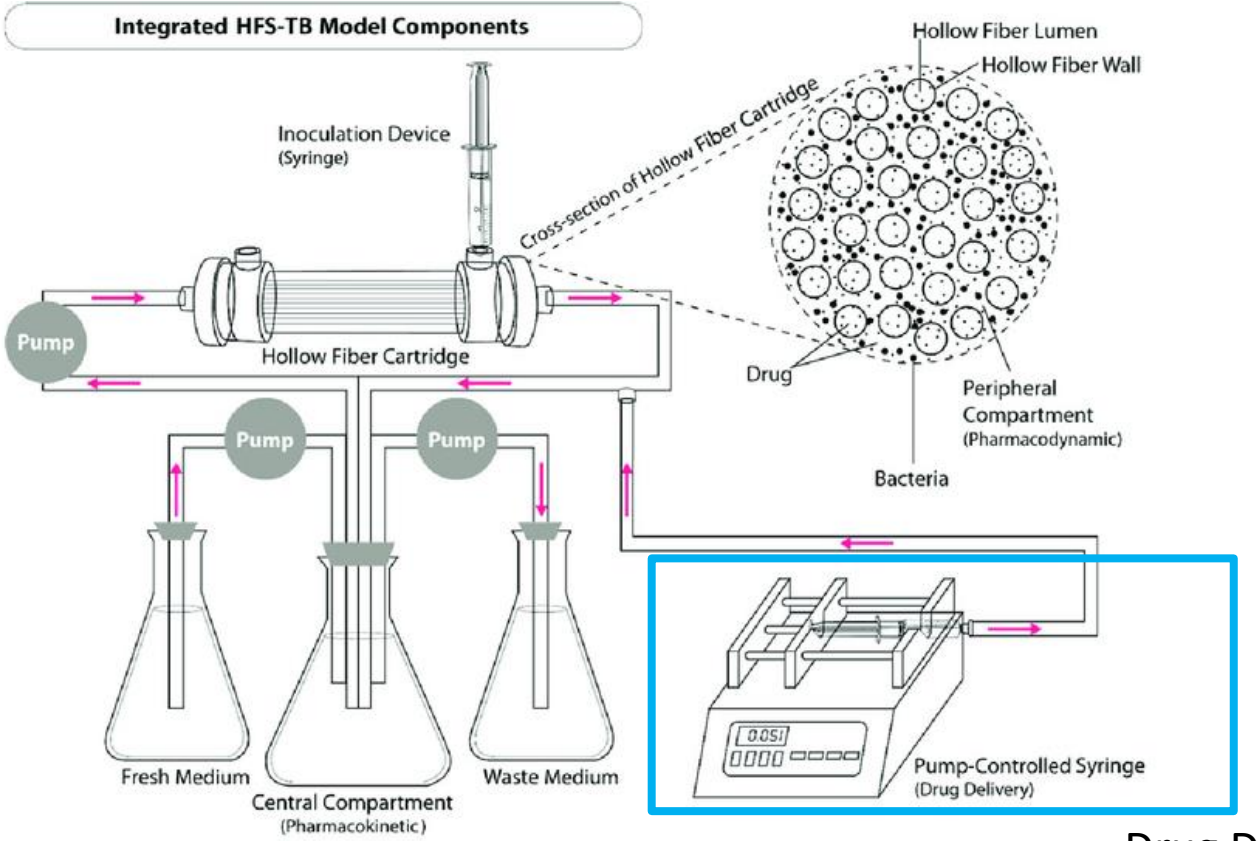
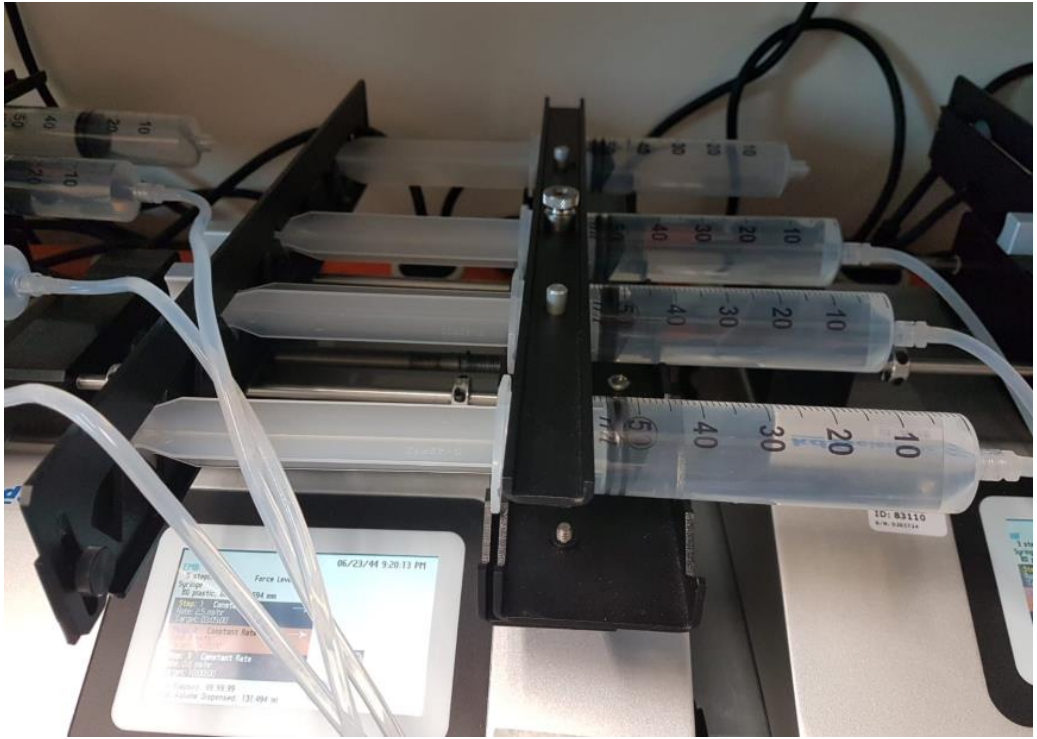


4L amber Jug (autoclavable)



Drug Delivery

Drug Delivery



kd Scientific Legato Series Computerized Syringe Pumps

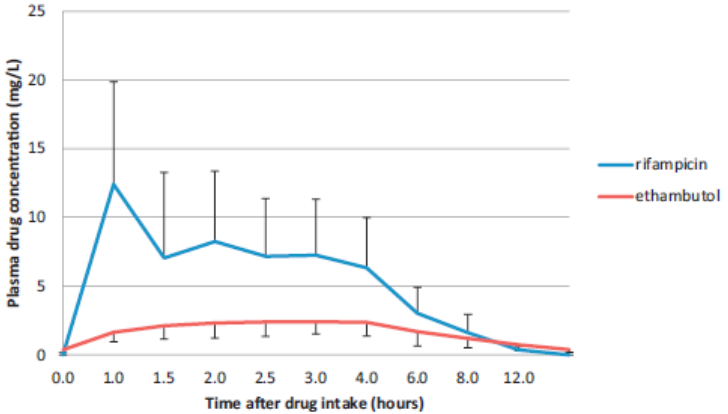
Multi-Step Programs to Accommodate for 0-order injection

60ml BD Plastipak LuerLock Syringes

Analysis using

Outcome

Readout



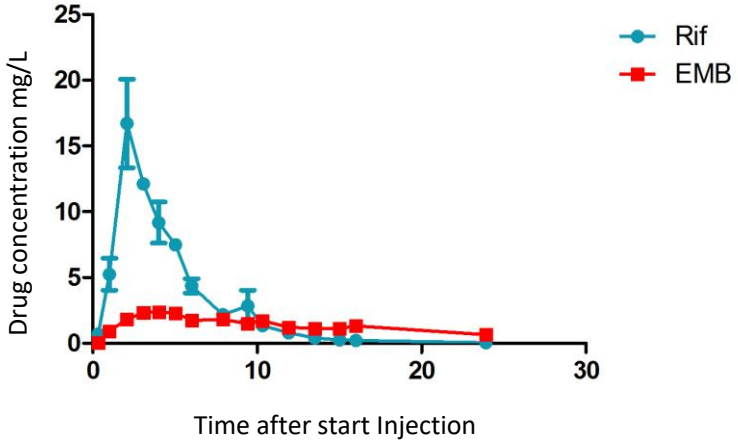


Fig. 1. Pharmacokinetics of rifampicin ($n = 14$) and ethambutol ($n = 13$). Data are the r

Pharmacokinetic parameter	RIF ($n = 14$)	EMB ($n = 13$)
AUC_{0-24} (RIF, EMB, AZM) or AUC_{0-12} (CLR, 14-OH-CLR) (h·mg/L)	45.9 (27.8–80.6)	24.2 (12.6–46.7)
C_{max} (mg/L)	12.3 (8.6–24.9)	3.1 (1.5–5.3)
T_{max} (h)	2.0 (1.0–4.0)	3.0 (1.5–6.0)
Cl/F (L/h)	13.1 (7.4–21.6)	46.5 (25.7–79.3)
V/F (L)	30.4 (16.5–45.2)	671.6 (338.6–1356.4)
$t_{1/2}$ (h)	1.6 (1.2–2.7)	10.0 (6.6–16.2)

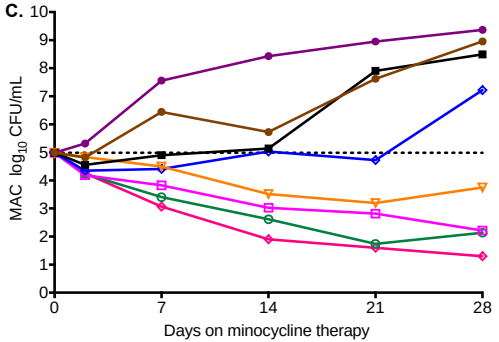
Parameter	Ruth
Auc 0-24	68.72
Cmax	16.71+/- 3.36
Tmax	2.0
T 1/2	2.2

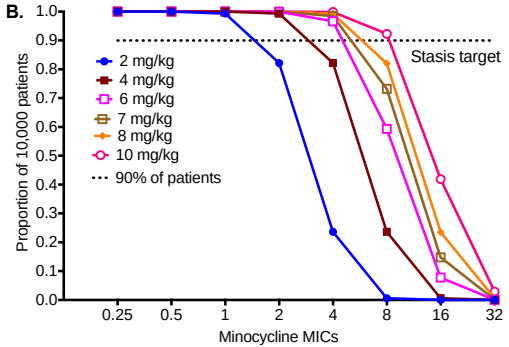
Parameter	Ruth
Auc 0-24	32.74
Cmax	2.31 +/- 0.3
Tmax	3
T 1/2	11

Magis-Escurra C. Int J Antimicrob Agents. 2013

Minocycline for MAC-PD

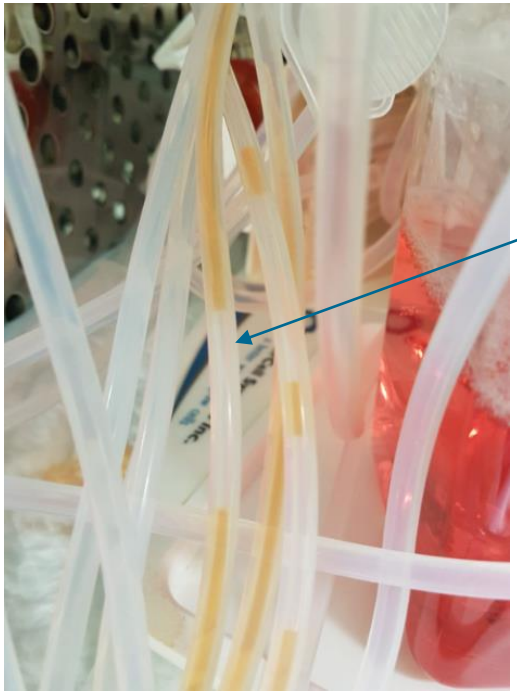
- Dose-Response HFS for MAC
- Monte-Carlo Based dose finding
- Identified 4mg/L as an effective dose





Ruth MM. J Antimicrob Chemother. 2019

Troubleshooting

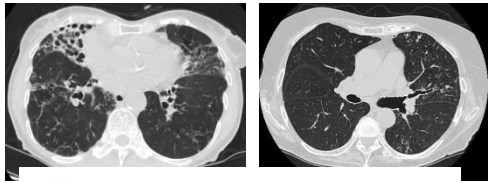


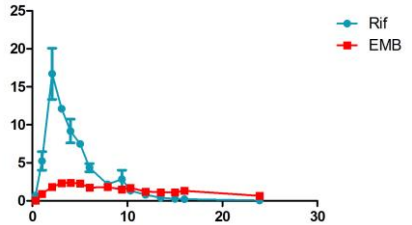
Back-tracking of drugs in tubing
Over night

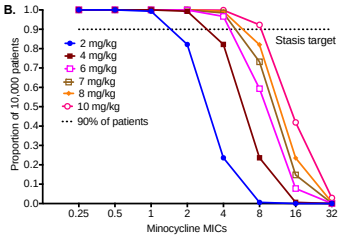
Summary: PK/PD and NTM-PD therapy design

- PK/PD has not been used much in NTM treatment
- Current MAC-PD regimen mostly ineffective (RIF! EMB!)
- We aim to develop a pk/pd informed new treatment regiment

Using the hollow-fiber model







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Radboudumc

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Reinout van Crevel

Pulmonary Diseases

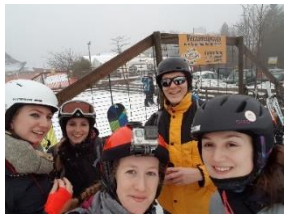
Wouter Hoefsloot

Martin Boeree

Cecile Magis

Sanne Zweijpfenning

Mycobacteriology team



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