Pre-clinical tools for evaluating new components of TB regimens

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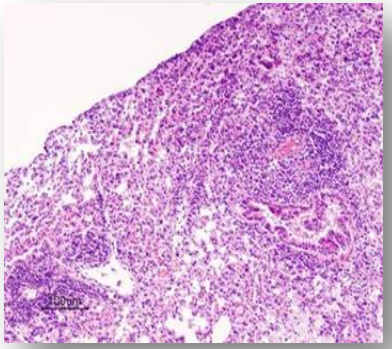
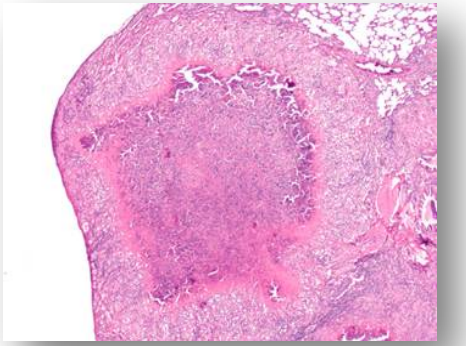
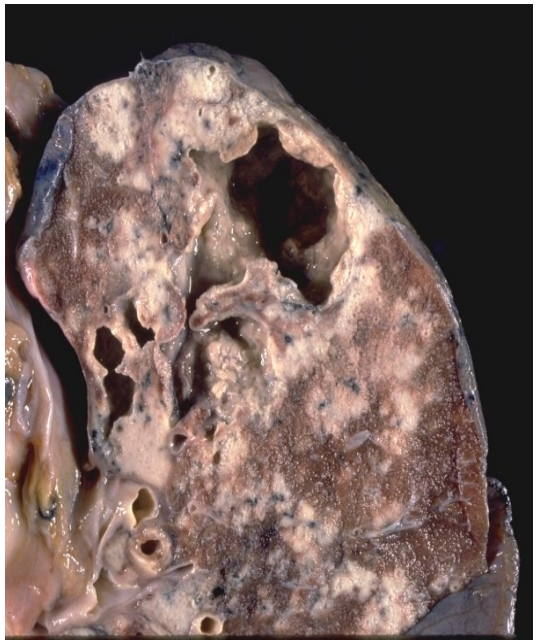
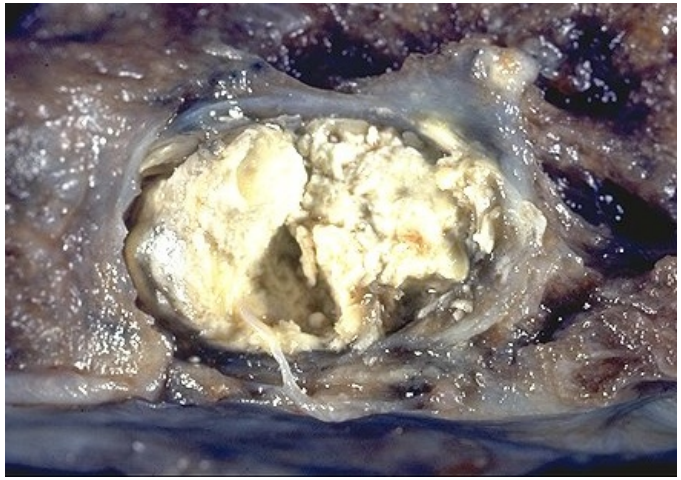




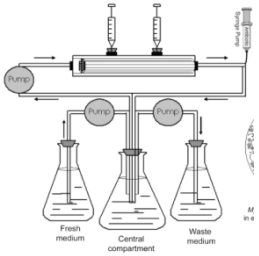
"All models are wrong,
some are useful."

George Box





In vitro and in vivo models
Pros & cons



In vitro models

Pros

- controlled microenvironment
- unlimited range of doses, schedules
- more precise measurement of drug conc. to which *Mtb* is exposed
- simple, serial sampling of *Mtb* products

Cons

- difficult to account for host effects on lesion microenvironment, microbial growth and susceptibility, and drug exposures at site of infection

In vivo models

Pros

- embody dynamic interaction b/w host, drug and microbe & represent impact of pathology
- enable simultaneous study of multiple sub-populations, perhaps in clinically relevant proportions

Cons

- limitations in dose size and schedules
- often difficult to mimic human PK
- may or may not represent diverse human disease states well

EMA qualification opinion on the HFS-TB

June 26, 2014

- HFS-TB qualified for use in drug development programs ***as an additional and complementary tool***
- HFS-TB can be used in regulatory submissions, esp. for informed design and interpretation of clinical studies
- HFS-TB is recommended to be useful as follows:
  - To provide preliminary proof of concept for developing a specific drug or combination to treat tuberculosis
  - To select the pharmacodynamic target (e.g. $T_{>MIC}$, AUC/MIC)
  - To provide data to support PK/PD analyses leading to initial dose selection for non-clinical and clinical studies
  - To assist in confirming dose regimens for later clinical trials taking into account human PK data and exposure-response relationships

Some unanswered questions for HFS-TB

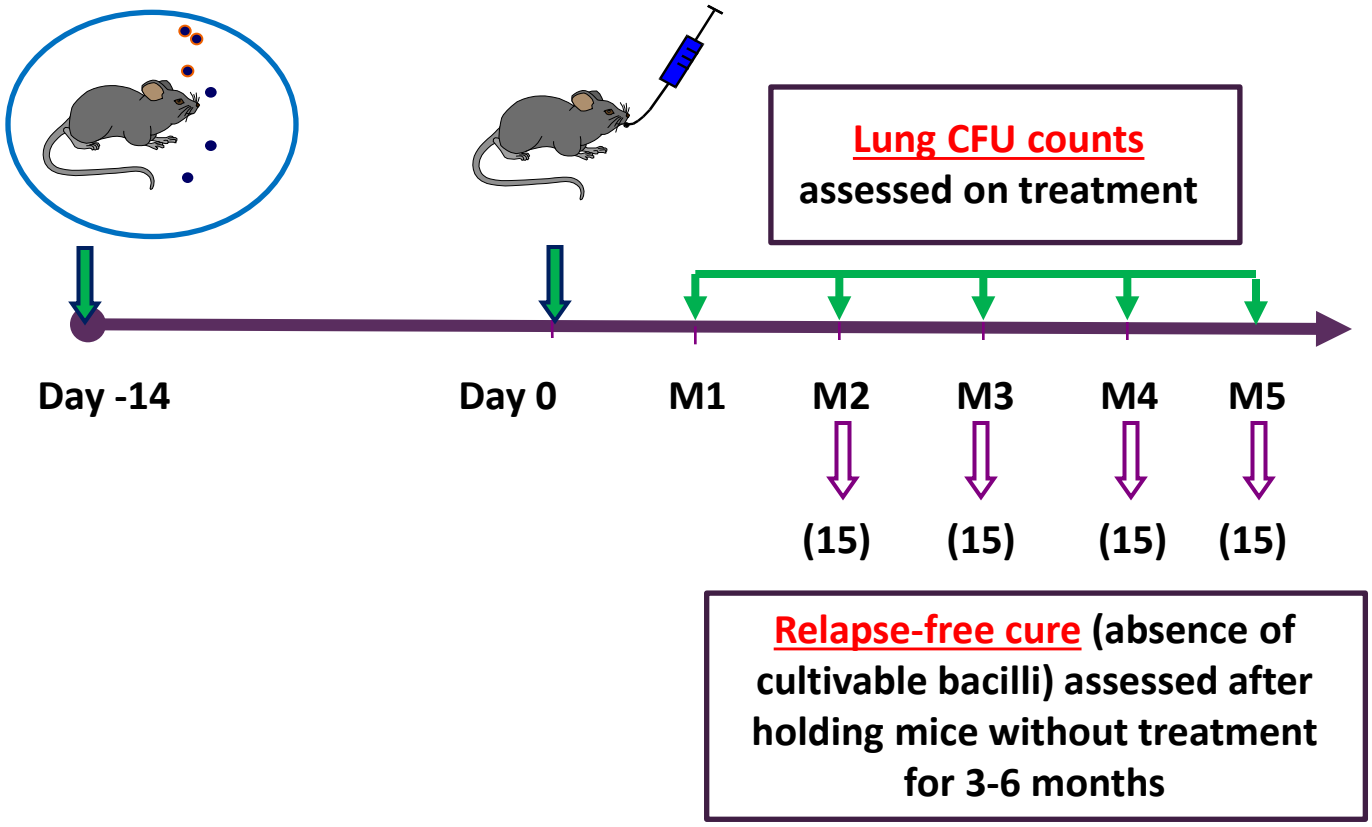
- Reproducibility?
- Obstacles to technology transfer and uptake?
- Reliability of estimates of drug exposures at site of infection (eg, using free drug fraction, ELF penetration ratios)?
- Predictive accuracy for efficacy of regimens?
  - rank ordering existing and novel regimens
  - estimating absolute or relative treatment durations
- Optimal integration of log phase and sterilizing effect models to predict regimen efficacy?

Caveat

"Correlations between drug concentration and pathogen survival that are based on in vitro models cannot be expected to reiterate all aspects of in vivo antimycobacterial treatment."

Chilukuri et al, CID 2015; 61(S1):S32

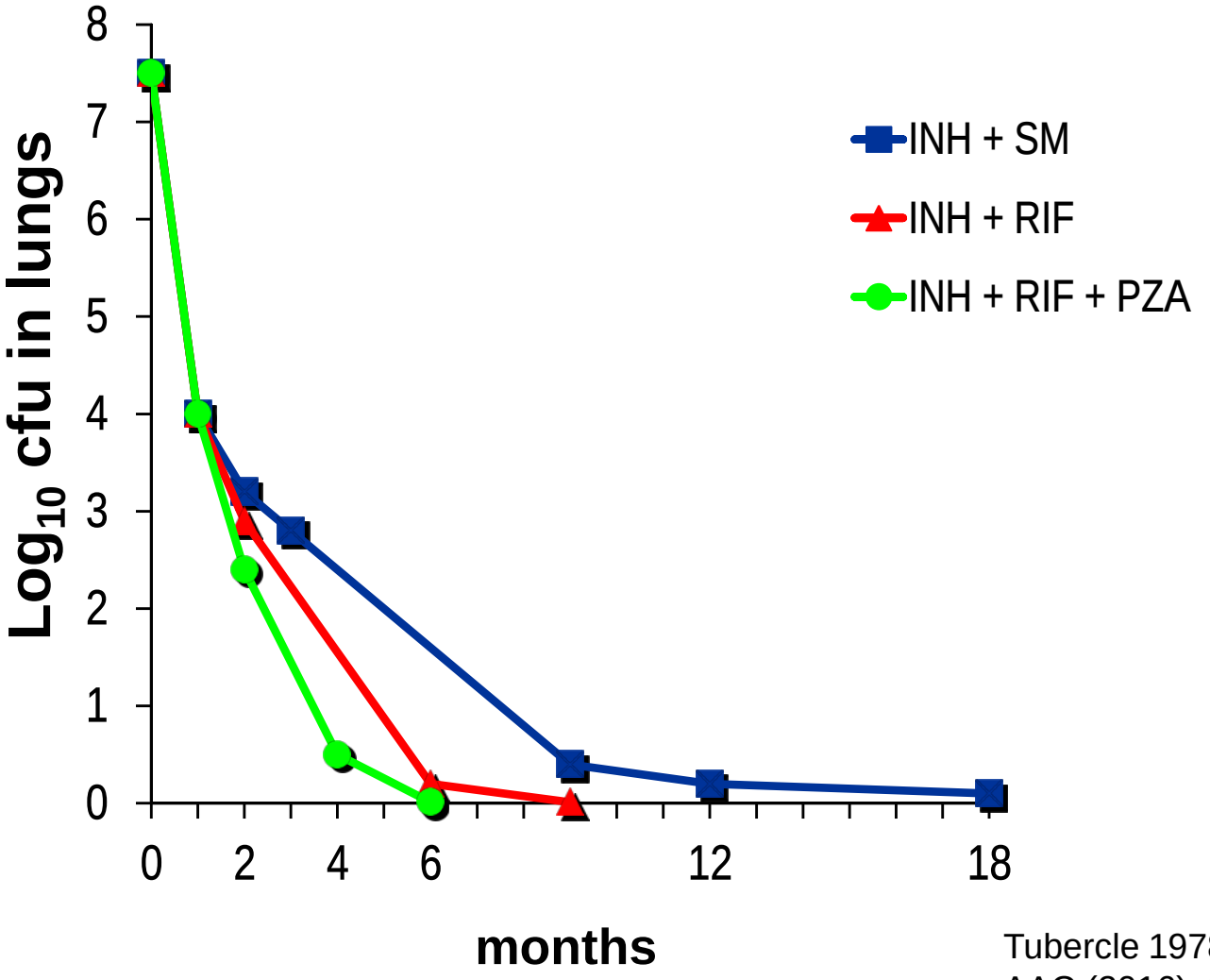
Scheme for relapse-based experiments in mice



Current uses of mouse models in the context of TB regimen development

- Derive (or confirm) PK/PD relationships for selecting optimal doses of component drugs
- Rank order drug combinations on the basis of efficacy
- Estimate treatment-shortening potential
- Assess impact of caseous pathology (eg, in Kramnik mice)
- Estimate potential for selection of drug-resistant mutants

Recapitulation of the short-course regimen in the mouse…as in humans



Tubercle 1978: 59:287 & 1986;67:5
AAC (2016); 60:1091

Performance of novel regimens in BALB/c mice (HDA model)

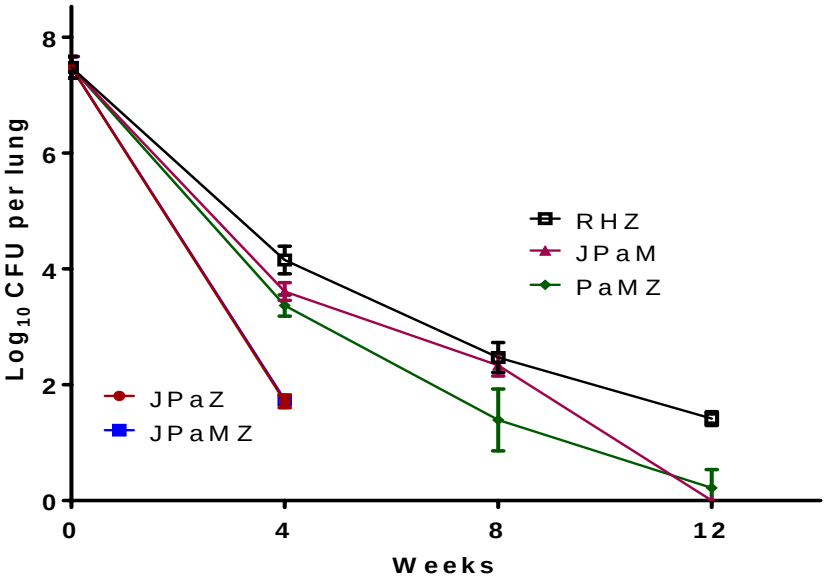
Regimen	Treatment-shortening effect (in months) relative to RHZ(E)
RMZ(E)	1 - 1.5
PaMZ	0 - 1
BPaMZ	3 - 3.5
BPaL	1-1.5 months

Pa = pretomanid; M = moxifloxacin; Z = pyrazinamide; B = bedaquiline; L = linezolid

- The PCS working group of CPTR has embarked on an effort to quantify the predictive accuracy of the “sterilizing” mouse model
- New regimens in, or advancing to, phase 3 trials will provide additional opportunities to evaluate the correspondence

Contribution of component drugs to the efficacy of the BPaMZ regimen

Bactericidal effect



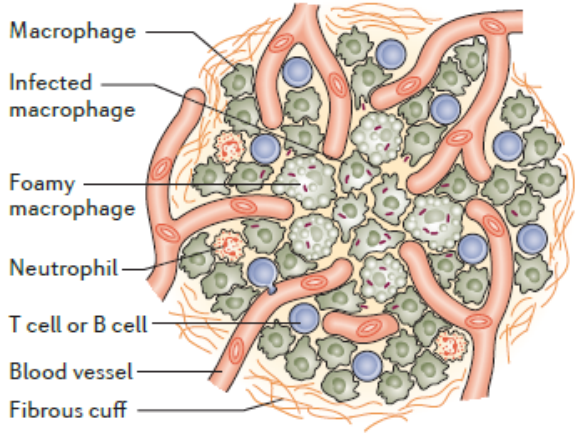
Sterilizing effect

	Proportion of mice relapsing by time point:				
Regimen	M1.5 (+3)	M2 (+3)	M3 (+3)	M4 (+3)	M5 (+3)
RHZ				10/15	2/15
PaMZ			10/14	3/15	
JPaM			2/15	0/14	
JPaZ	13/14	0/15	0/15		
JPaMZ	3/15	0/15	0/15		

Li et al, AAC 2017; in press

Challenges in translating mouse model results to the clinic

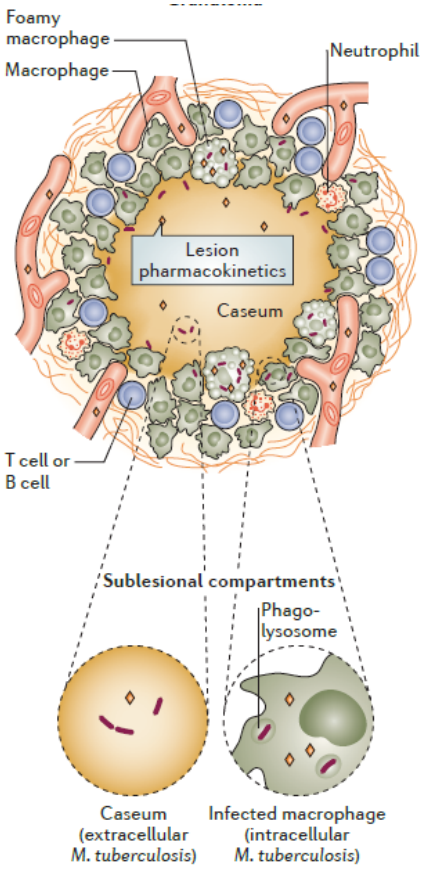
Cellular granuloma
BALB/c, C3HeB/FeJ mice



Differences in host response and lung pathology

Differences in:
Mtb growth rate
Intra/extracellular residence
Drug distribution
Lesion microenvironment

Caseating granuloma
C3HeB/FeJ mice only



V. Dartois, Nat Rev Microbiol, 2014

Examples of different outcomes in C3HeB/FeJ mice compared to BALB/c mice

- Lack of pyrazinamide bactericidal effect in large caseous lesions, where caseum has neutral pH[1]
  - yet, addition of PZA to RIF+INH+EMB still shortens treatment duration[2]
- Lack of clofazimine bactericidal effect in large caseous lesions, where CFZ diffuses poorly and caseum has neutral pH and is hypoxic[3]
- Reduced bedaquiline effect in large caseous lesions, where BDQ diffuses poorly[4]

1. Lanoix et al, AAC 2016; 60:735
2. Lanoix et al, AAC 2016; 60:1091
3. Irwin et al, AAC 2014; 58:4026
4. Irwin et al, ACS Infect Dis 2016; 2:251

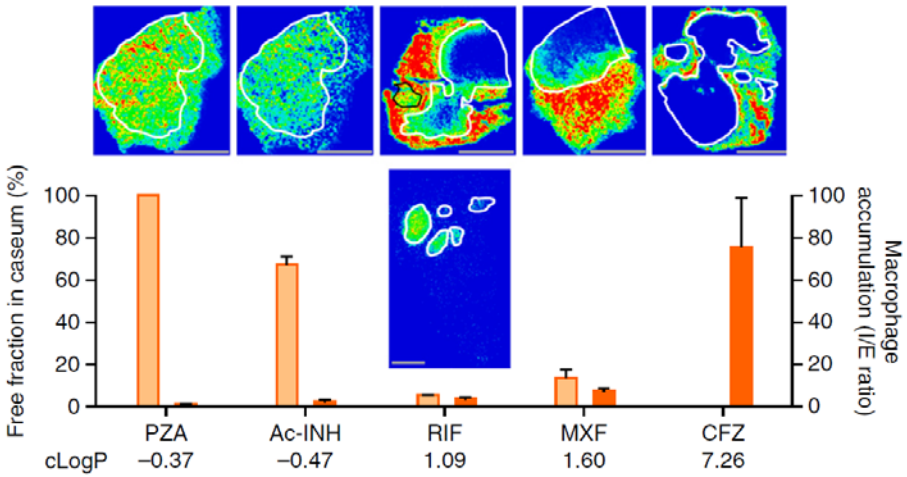
Limited experience comparing regimens in other animal models with caseous pathology

- **Guinea pigs**
  - Replacing RIF with RPT had no significant treatment-shortening effect[1]
  - PaMZ had no significant treatment shortening effect compared to RHZ[2]
- **Rabbits**
  - No regimen comparisons found
- **Marmosets**
  - RHZE reduced the extent of disease on PET-CT and lowered CFU counts in cavities compared to HS after 6 weeks of treatment[3]
- **Macaques**
  - Metronidazole did not increase the bactericidal effect of RH

1. Ahmad et al, AAC 2012; 56:3726
2. Dutta et al, AAC 2013; 57:3910
3. Via et al, AAC 2014; 59:4181

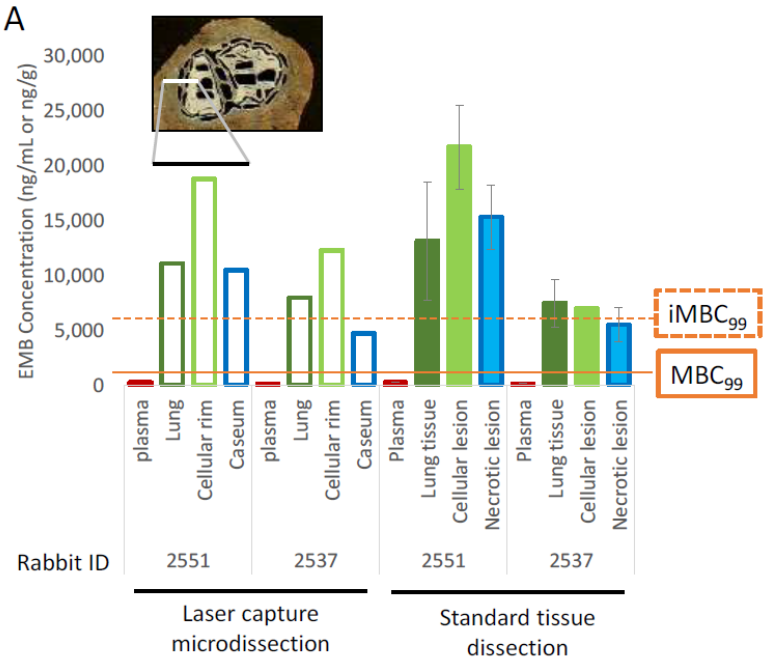
Evaluating drug partitioning into TB lesions

MALDI-MSI



B Prideaux et al, Nature Medicine, 2015

Laser-capture microdissection and LC/MS-MS



M Zimmerman et al, AAC, 2017

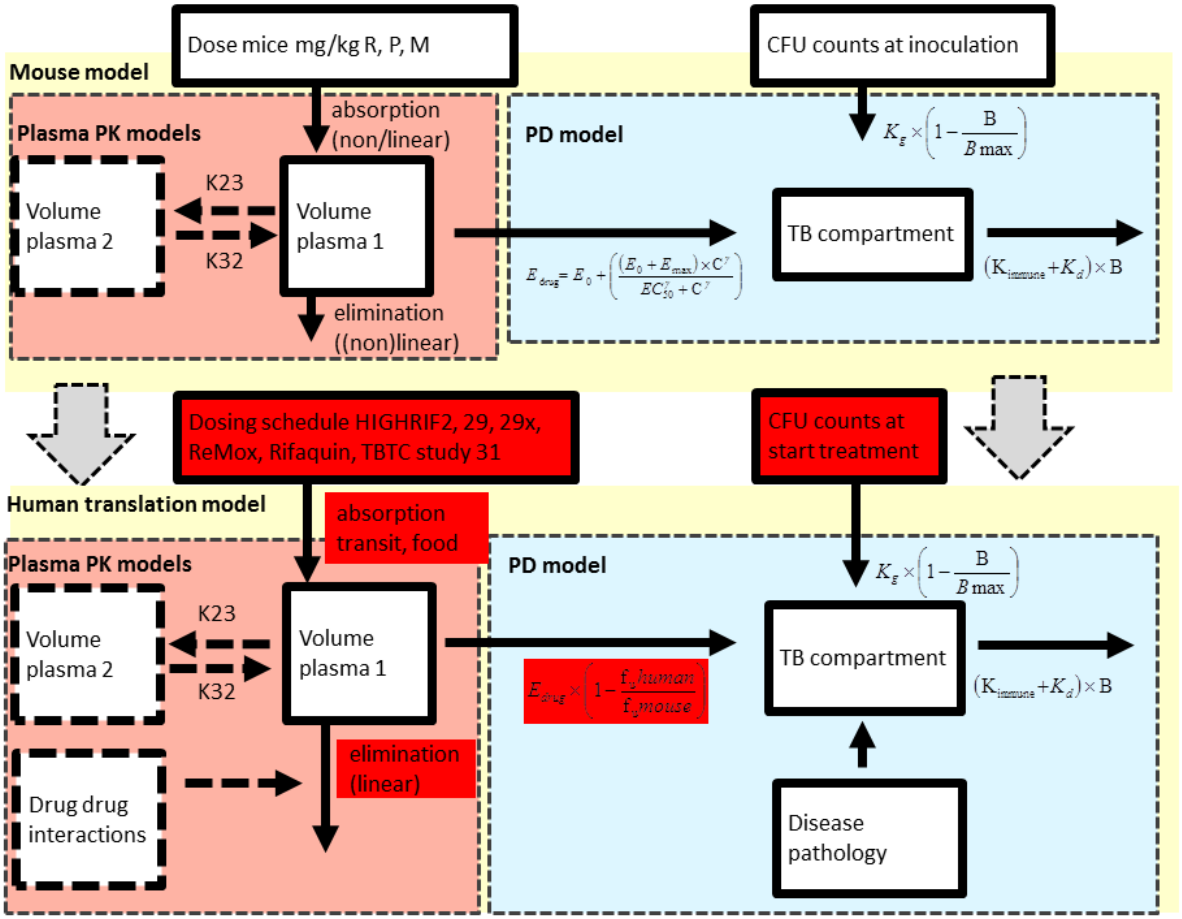
Additional challenges in translating mouse model results to the clinic

- Inter-species differences in drug PK
- Experiments in inbred mice infected with 1 *Mtb* strain and treated with identical drug doses cannot recapitulate the many sources of heterogeneity in human TB:
  - PK variability
  - Severity of disease (eg, presence of cavities, cavity size)
  - Immune status
  - Adherence to treatment
  - *Mtb* drug susceptibility

Translational (mouse → human) PK/PD Model
Objectives

- Develop a translational PK/PD model that utilizes:
  - mouse PK data for RIF, RPT and MXF
  - mouse PD data (CFU counts) for RIF, RPT and MXF alone and in combinations including PZA + INH or EMB
  - human PK data, including rifamycin-MXF interaction
  - an immune effect on bacterial death derived from CFU differences between nude and BALB/c mice
  - inter-species differences in protein binding
  - effect of caseation and cavitation on lesion distribution of rifamycins
- Perform clinical trial simulations to predict trial outcomes
  - sputum culture status at 8 wks
  - relapse status at 1 yr

Translational (Mouse → Human) PK/PD Model



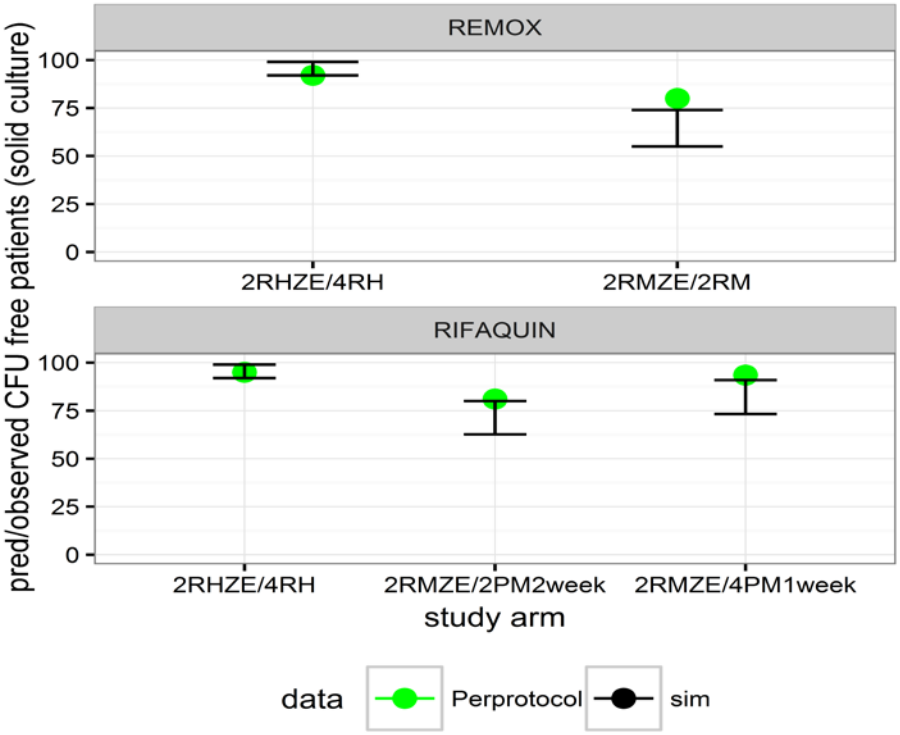
R= rifampin
P= rifapentine
M= moxifloxacin

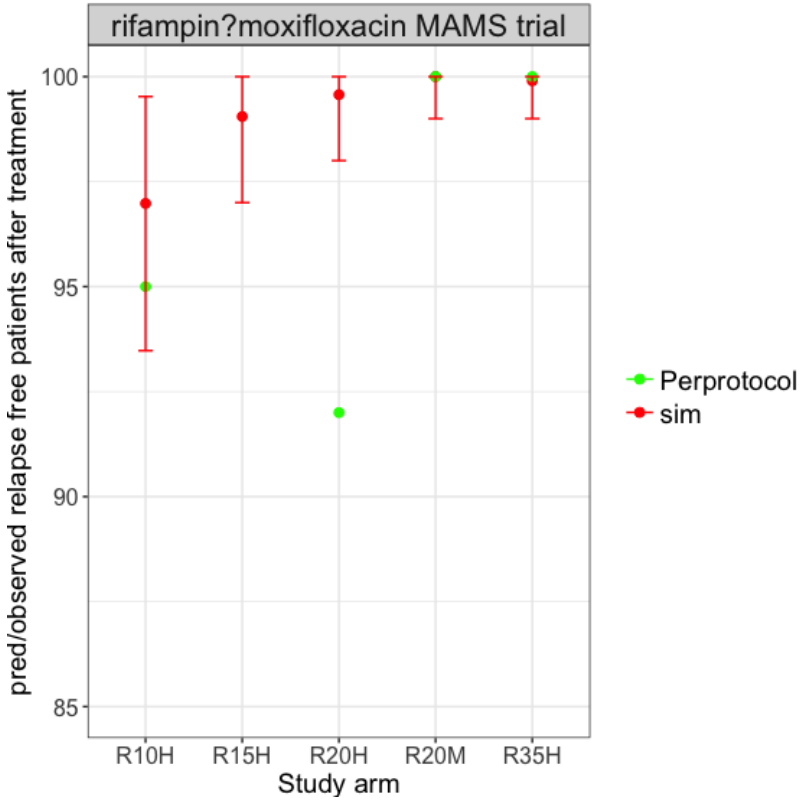
B_{MAX}= maximum number of bacteria
K_{growth}= bacterial growth constant
K_{death}= bacterial death constant
IT_{50}= time of 50% of max. immune response
θ_{KIND}= max. immune kill rate (untreated mice)
$\gamma_{immune\ response}$ = sigmoidicity factor, defines shape of immune response effect
$\theta_{KDOI.0}$= immune kill rate (treated mice) at average incubation time
$\theta_{KDOI.t}$= increase in kill rate (treated mice) in expts w/above average incubation time
E_{drug} = drug effect
E_{max}= max. achievable drug effect
EC_{50} = antibiotic conc. producing 50% of E_{max}
γ = sigmoidicity factor, defines the shape of drug effect

Bartelink et al, Clinical and Translational Science, In press

Using the final translational PK/PD model:

Predicted vs. observed trial results





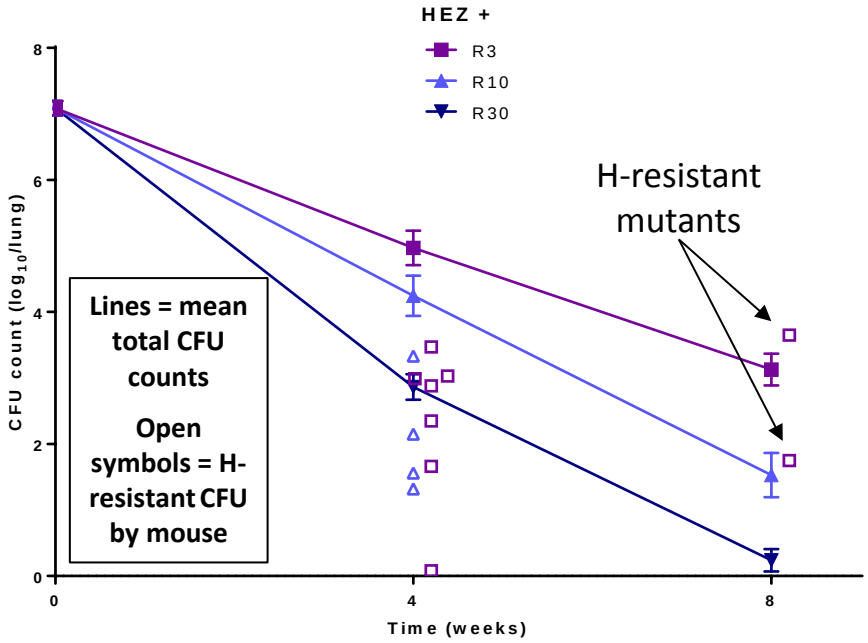
Bartelink et al, Clinical and Translational Science, In press

Translational PK/PD Model - conclusions

- The model performed reasonably well, especially in predicting higher relapse rates for 4-month arms
- Work is ongoing to:
  - incorporate individual PK/PD and dose-response for all drugs in regimens
  - simulate phase 3 trials with more novel regimens
  - incorporate drug-resistant sub-populations to predict rates of resistance emergence
  - merge PK/PD model with mechanistic within-host model to gain greater insight into factors driving regimen performance

Assessing the risk of resistance amplification

Impact of simulated RIF PK variability in C3HeB/FeJ mice



Low R exposures lead to acquired INH resistance in 2 of 10 mice

Impact of intermittency and immunodeficiency in nude mice

Recapitulating the arms of TBTC Study 22, selection of AHR and ARR were associated with:

- immunosuppression
  - nude mice more likely than BALB/c to have AHR (8.5% vs 0%, p= 0.001) and ARR (3.5% vs. 0%, p= 0.06)
- intermittent vs daily initial phase therapy
  - 30% vs 2.7% for AHR/ARR (p< 0.001)
  - 20% vs 2.7% for ARR only (p< 0.01)
- once-weekly RPT vs RH in contin. phase
  - 18% vs 3.3% for ARR (p< 0.05)

Park et al, submitted

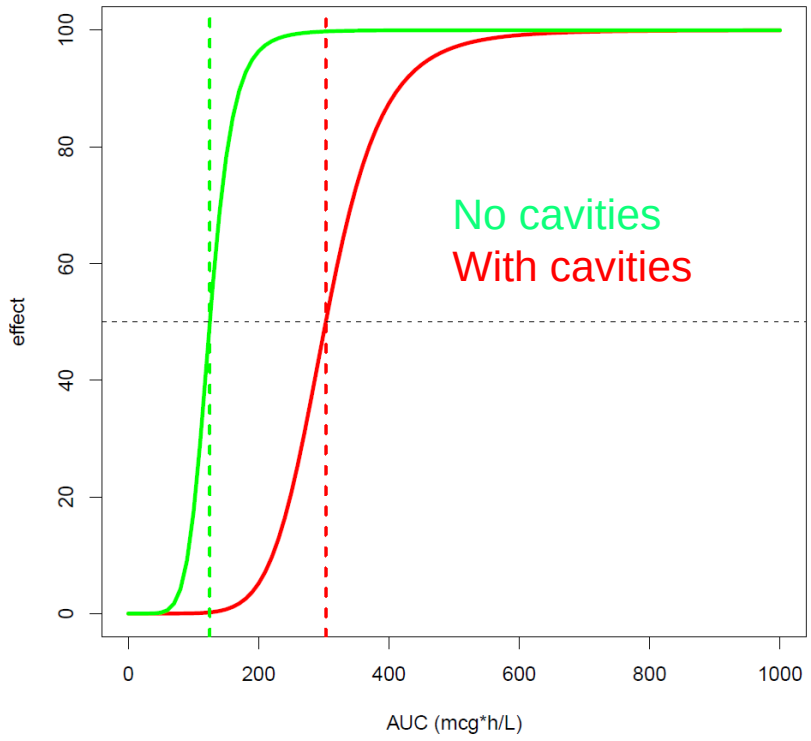
Take-home points

- *In vitro* hollow fiber models are qualified as useful tools for exploring PK/PD relationships under controlled conditions
- Mouse models have an established track record in estimating the treatment-shortening potential of novel regimens
- The impact of certain variables that modify the effect of some drugs may require elucidation in caseous disease models:
- Emerging data from clinical trials with novel regimens will provide a great opportunity for further evaluating the predictive accuracy of these and other preclinical models
- Some factors are more difficult to account for in pre-clinical models and may be best address with more predictive PK/PD-based translational models:
  - inter-species PK differences in PK, protein binding, etc
  - human PK variability
  - heterogeneity in human host (eg, cavitation, immune response)
  - heterogeneity in bacterial pathogen (eg, MIC distribution)

Acknowledgements

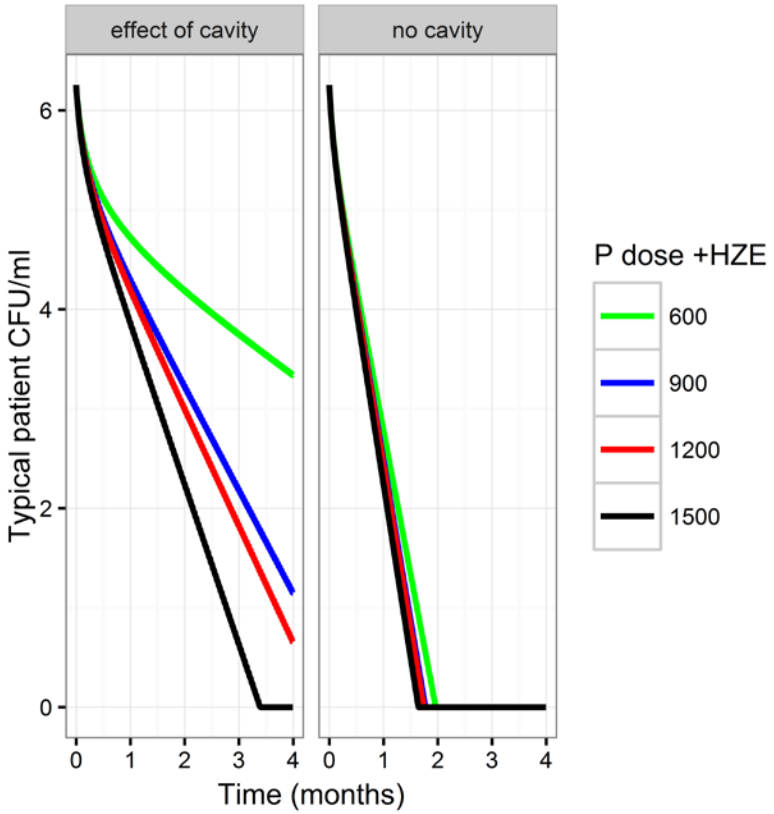
- Members of the lab and the JHU Center for TB Research
- Collaborators
  - Veronique Dartois (Rutgers)
  - Tawanda Gumbo (Baylor)
  - Debra Hanna and CPTR PCS-WG
  - Anne Lenaerts, Scott Irwin (CSU)
  - Chuck Peloquin (UF)
  - Rada Savic, Imke Bartelink, Nan Zhang (UCSF)
- Funding
  - NIAID (R01-AI090820, R01-AI111992)
  - Global Alliance for TB Drug Development
  - FDA (U18-FD004004)
  - Gates Foundation (TBDA #42581, OPP1037174)
  - C-Path
  - CDC TB Trials Consortium

Incorporating the effect of disease pathology into the model



A 4-fold higher EC_{50} for P in pts with cavitary disease.

Savic, R.M. *et al.* CPT 2017 Jan. 25

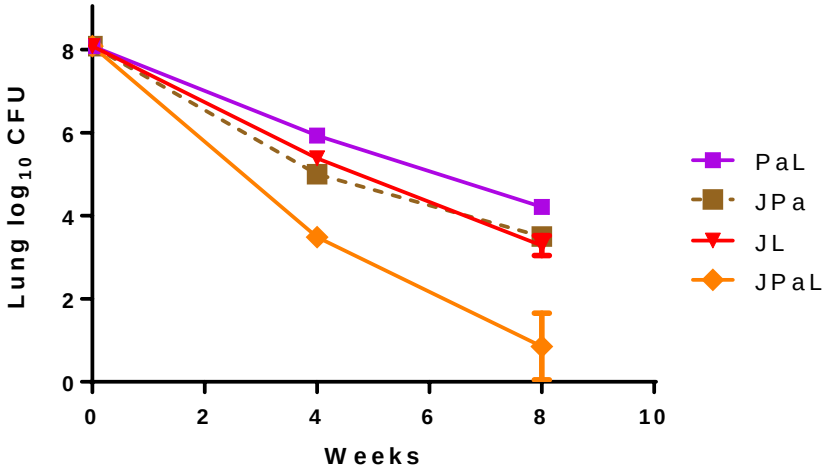


A 4-fold higher EC_{50} for P in the cavity compartment compared to plasma was used in the simulation.

Bartelink, I. *et al.* CPT 2017

Contribution of each component to the efficacy of the BPaL regimen

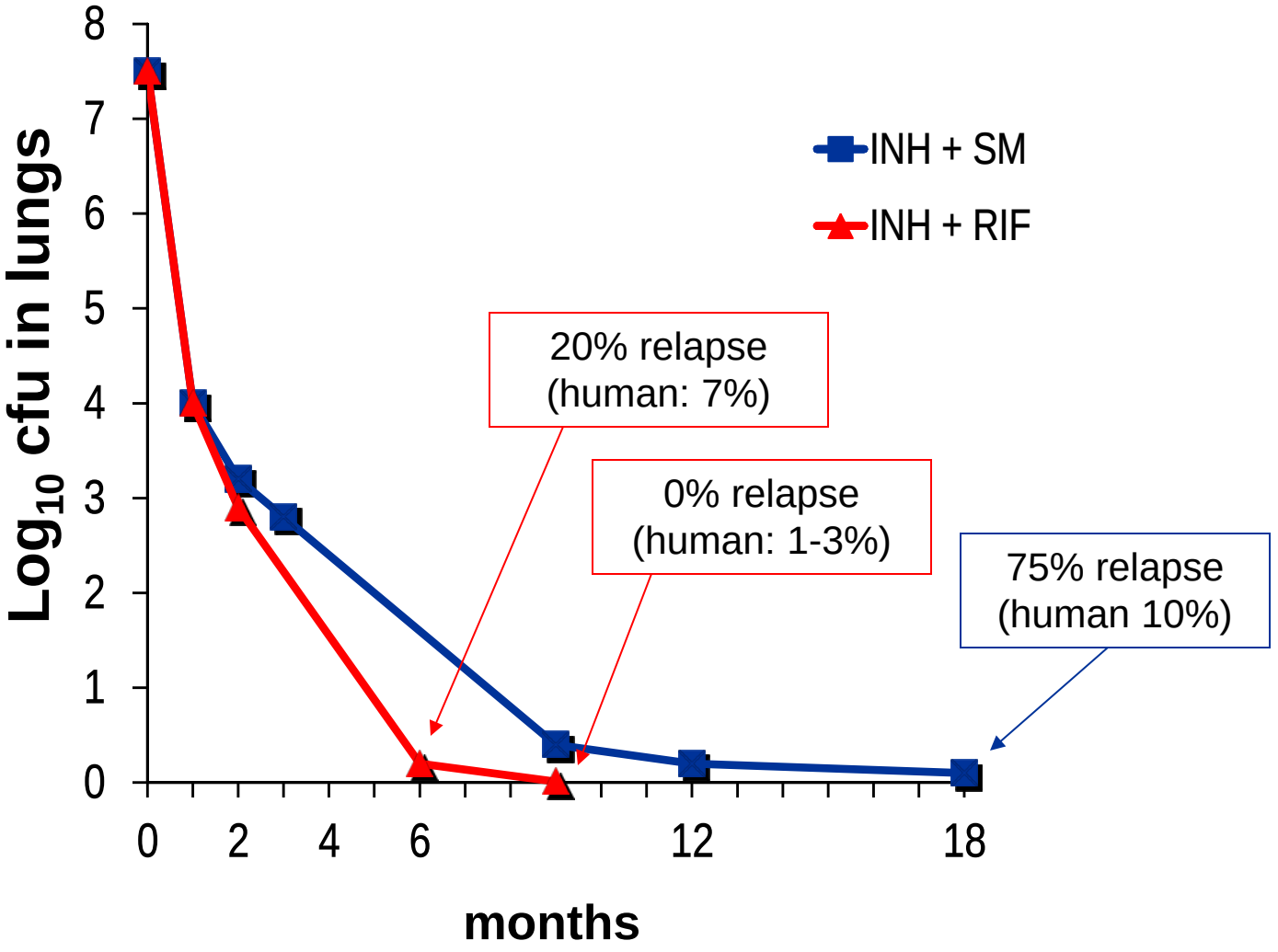
Bactericidal effect



Sterilizing effect

	Proportion relapsing after treatment for:	
Regimen	**2 months**	**3 months**
2RHZ/RH		**8/14**
JPa		**3/14**
JPaL		**0/15**
2JPaL/1JPa	**6/15**	**0/15**
1JPaL/2JPa	**9/15**	**0/15**

Recapitulation of the short-course regimen in the mouse…as in humans



Tubercle 1978;59:287 & 1986;67:5

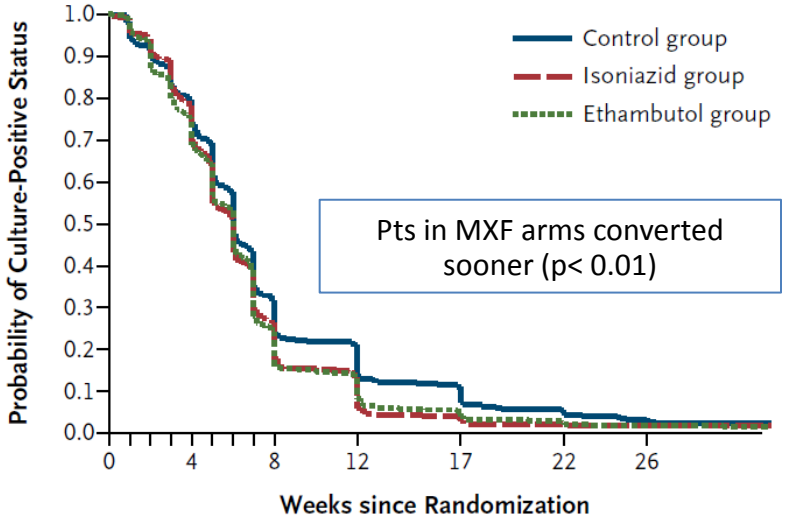
Recapitulating the evolution of short-course therapy in mice and humans*

Regimen	Months	Proportion Relapsing after Treatment:	
		Mice	Humans
INH+SM	6	100%	29%
INH+SM	18	75%	~10%
INH+RIF	6	20%	6-7%
INH+RIF	9	0-5%	1-3%
INH+RIF+PZA	4	70-90%	11-15%
INH+RIF+PZA	6	0-5%	1-3%

*From Mitchison; and Grosset & Ji; in Gangadharam & Jenkins, Chapman & Hall, 1998

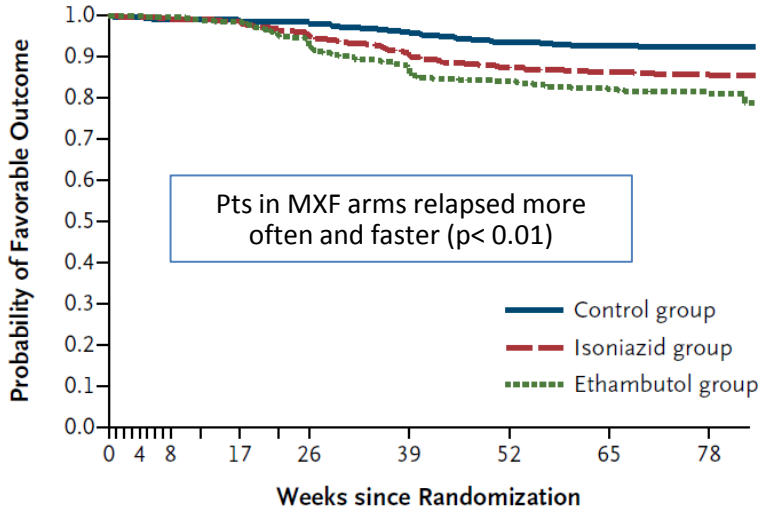
Moxifloxacin for treatment shortening: The REMoxTB phase 3 trial

Time to sputum conversion



No. at Risk						
Control	600	465	183	122	64	19
Isoniazid	617	459	154	76	21	9
Ethambutol	604	449	141	79	30	9

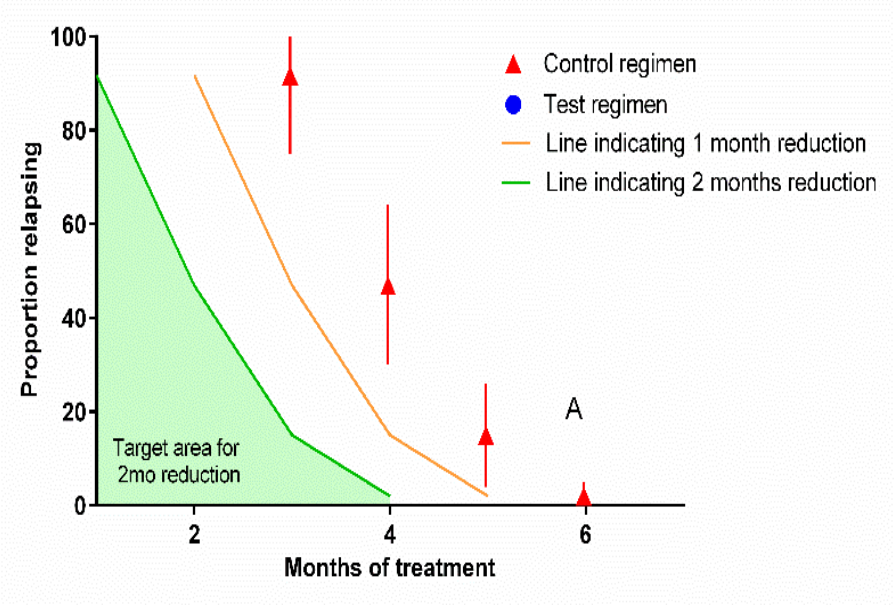
Time to unfavorable outcome

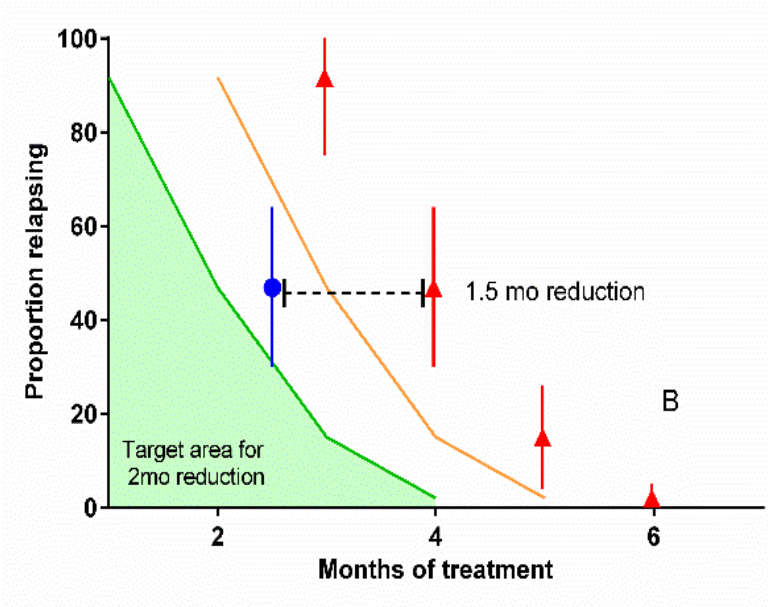


No. at Risk					
Control	600	563	533	493	472
Isoniazid	617	570	522	459	439
Ethambutol	604	568	523	445	425

Gillespie et al, NEJM 2014

Measuring the treatment-shortening effect of a test regimen relative to a control regimen in mice

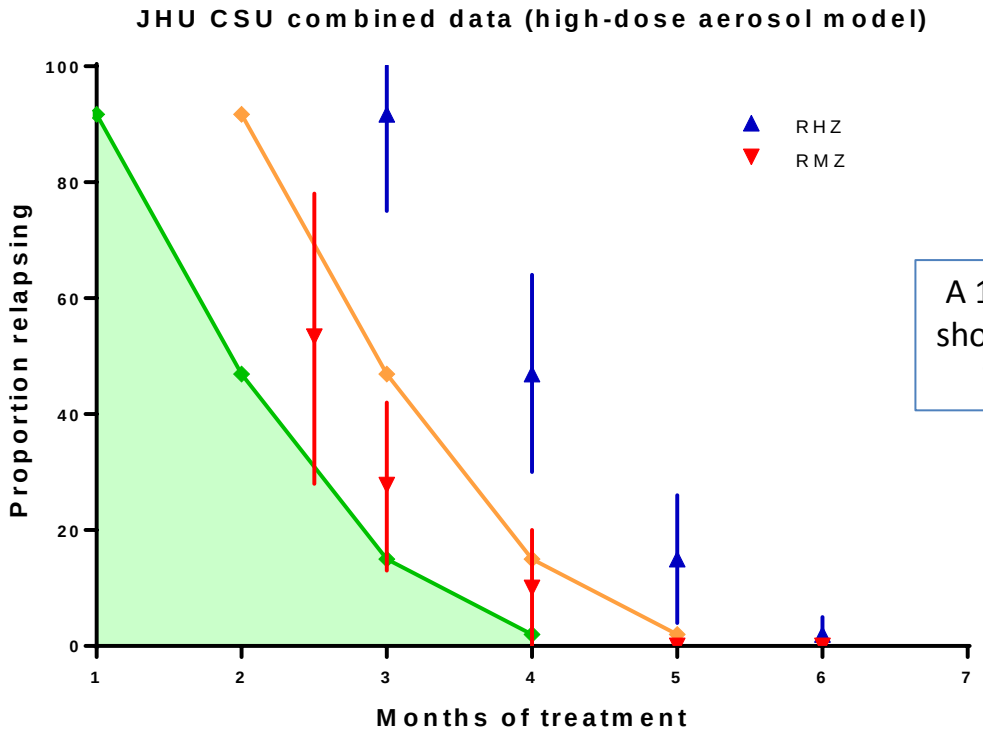




Colored symbols represent the proportion of mice relapsing after receiving the indicated regimen for various durations (error bars represent the 95%CI).

Lanoix et al, Clin Infect Dis 2016; 62:484

Treatment-shortening effect of substituting moxifloxacin for isoniazid in the 1st-line regimen in BALB/c mice



A 1 to 1.5-month treatment shortening effect is observed in our standard model.

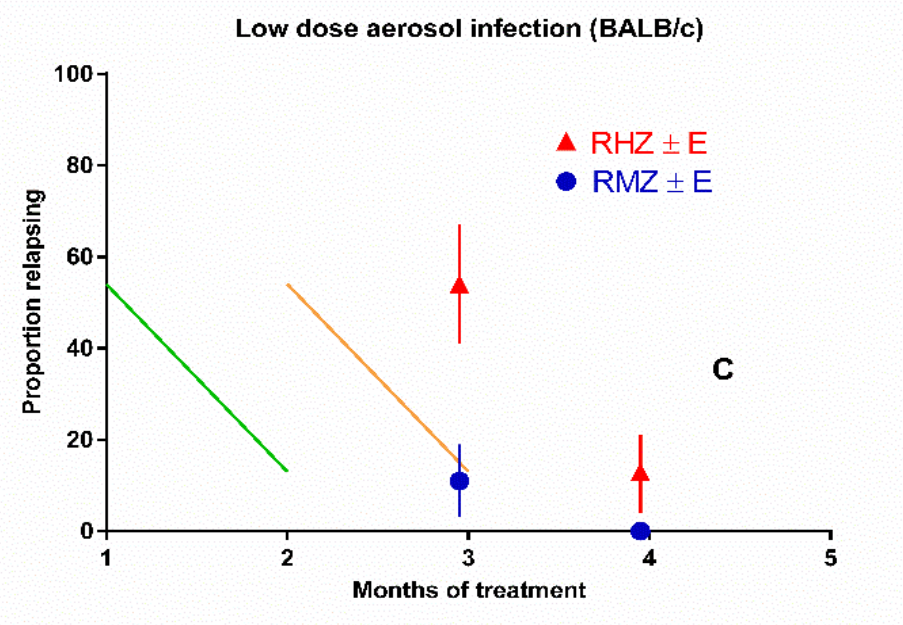
Data from 5 experiments (4 JHU, 1 CSU)

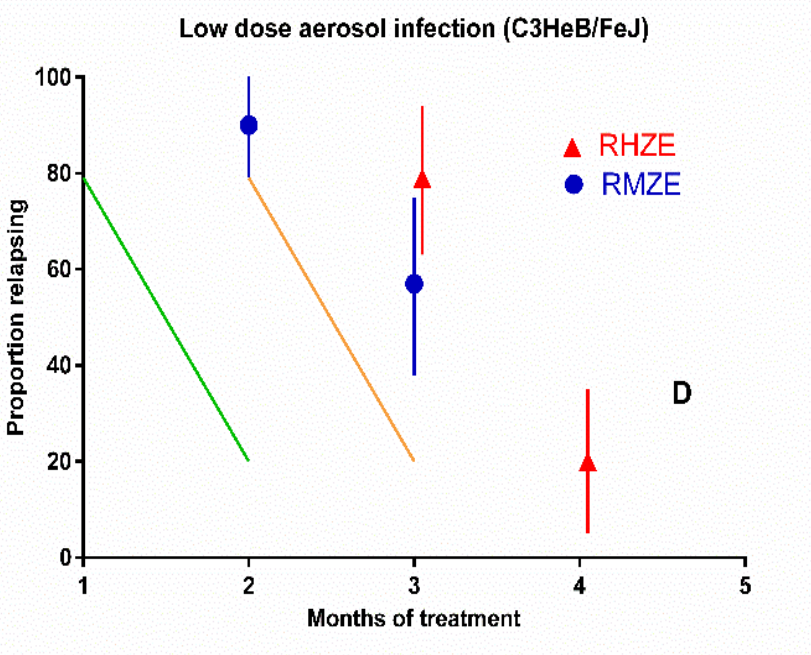
Lanoix et al, Clin Infect Dis 2016; 62:484

Outcomes with REMox-TB regimens in mice

- In REMox-TB, substitution of M for H or E resulted in faster sputum conversion but did not permit shortening the duration of treatment by 2 months
- In mice, **substitution of M for H** resulted in:
  - treatment shortening of 1-1.5 months in high-dose infection models
  - treatment shortening of 0-1 month in low-dose infection models
- In mice, **substitution of M for E** resulted in:
  - treatment shortening of 0-1 month in low-dose infection models
- Results in mice are not inconsistent with those of REMox-TB

Treatment-shortening effect of substituting moxifloxacin for isoniazid in the 1st-line regimen



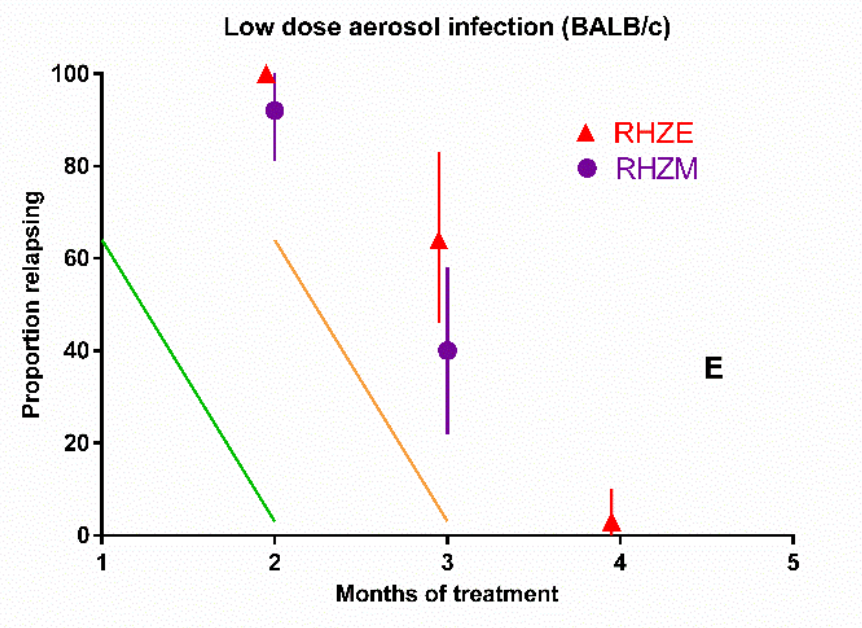


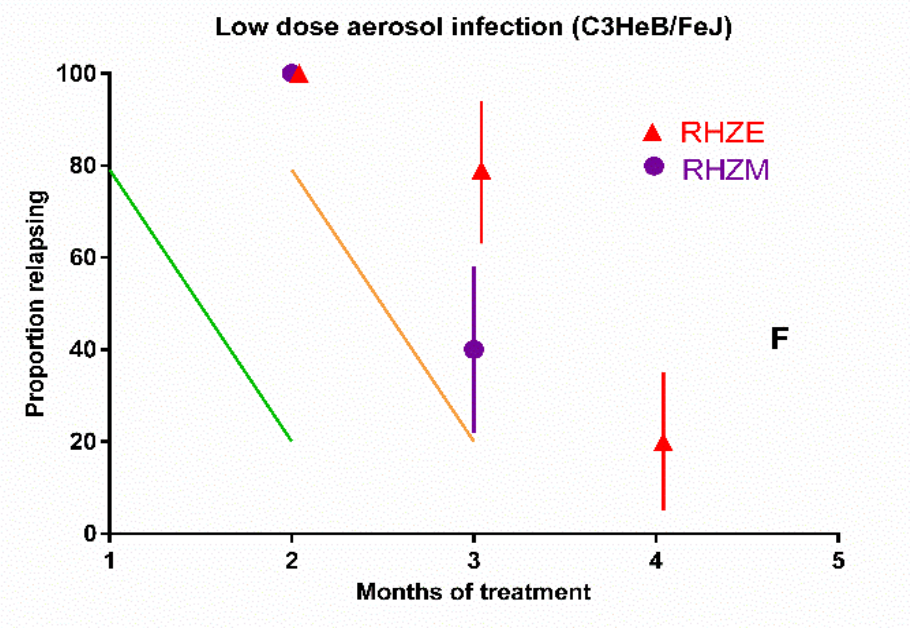
The treatment shortening effect is between 0-1 month in low-dose aerosol infection models in BALB/c and Kramnik mice.

Data from 2 experiments (1 JHU, 1 CSU) per mouse strain

Lanoix et al, Clin Infect Dis 2016; 62:484

Treatment-shortening effect of substituting moxifloxacin for ethambutol in the 1st-line regimen





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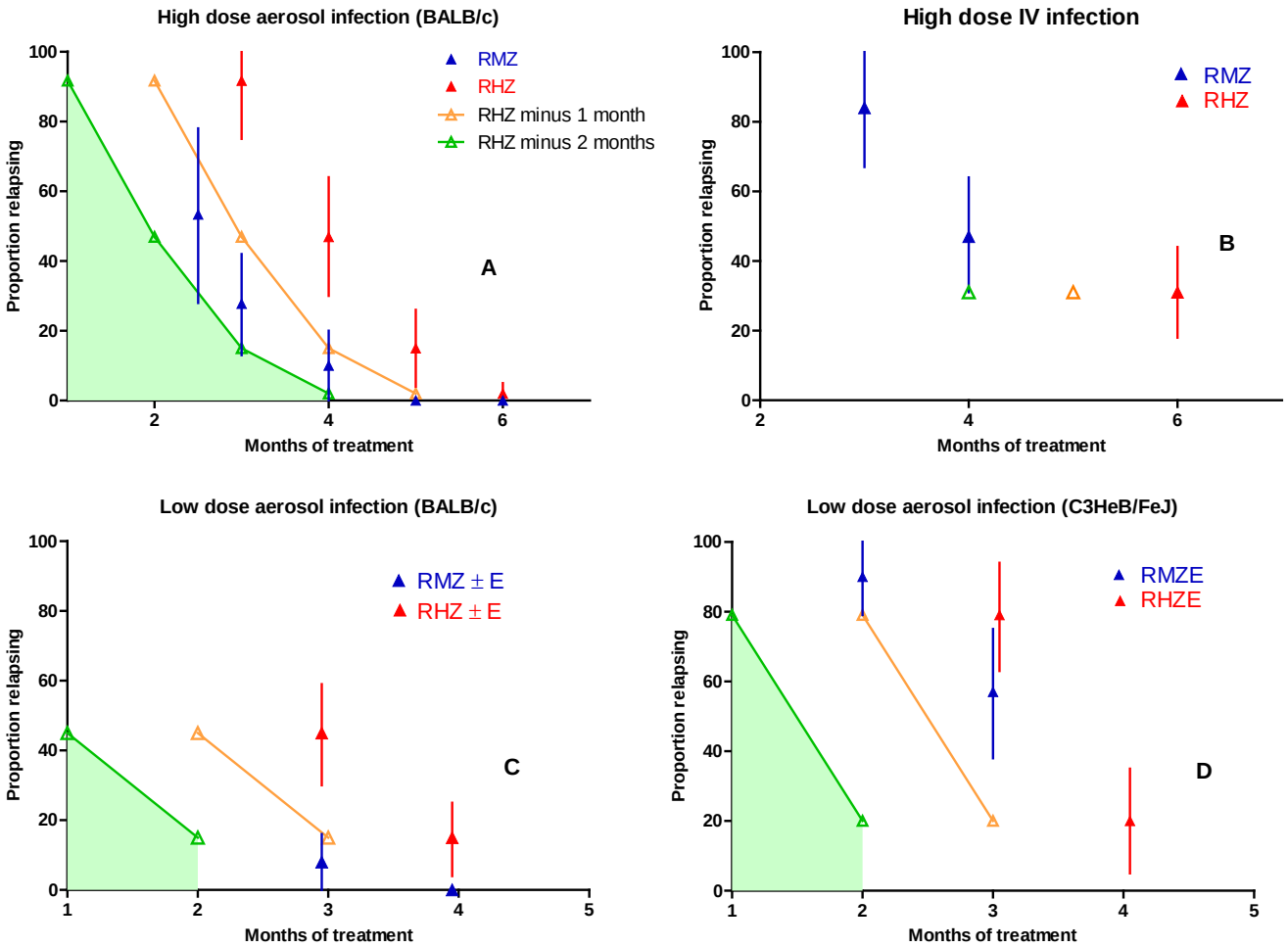
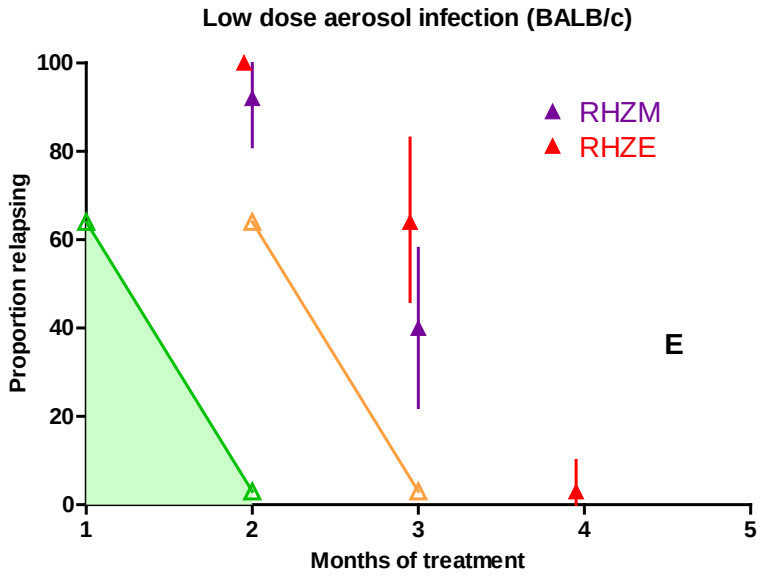
Data from 2 experiments (1 JHU, 1 CSU) per mouse strain

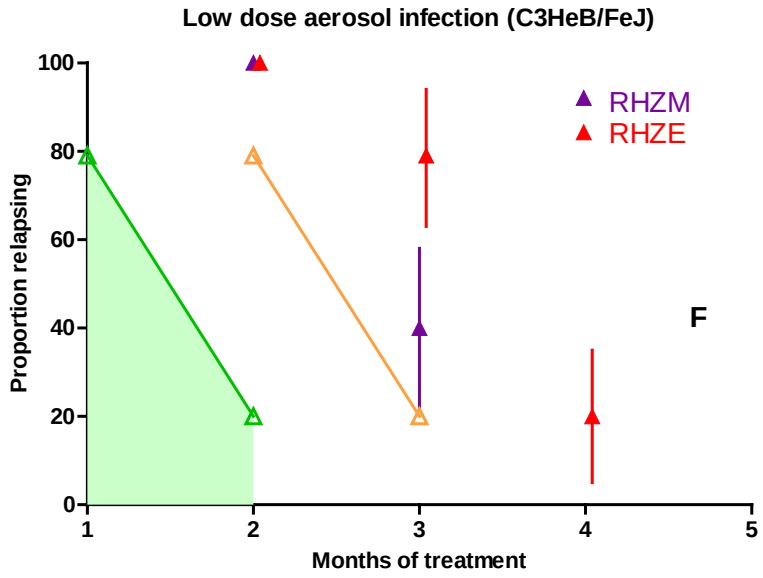
Lanoix et al, Clin Infect Dis 2016; 62:484

Substitution of M for H in the RHZ regimen in mice – data from 3 institutions, using low- & high-dose infection models

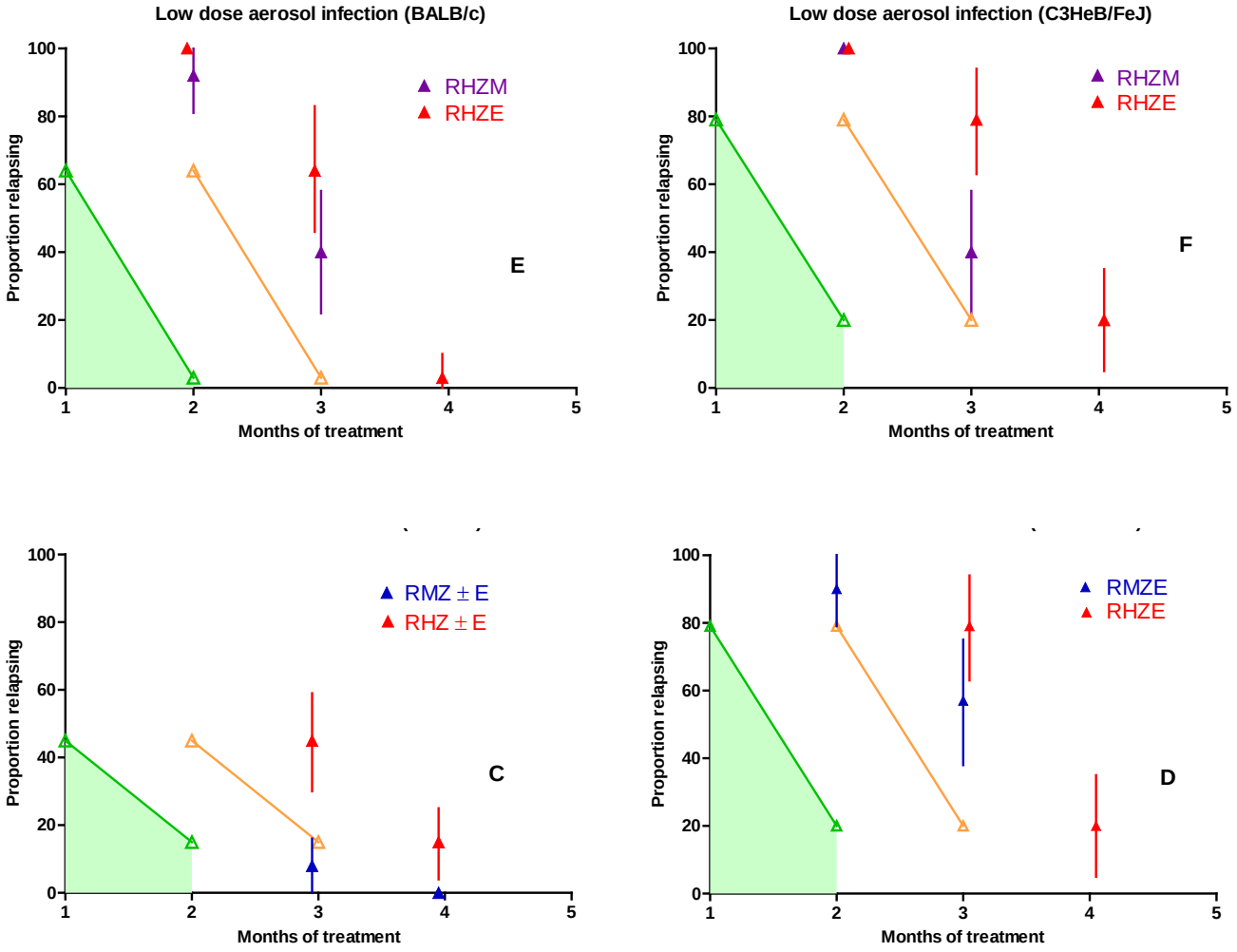


Substitution of M for E in RHZE – data from 2 institutions in chronic infections in 2 mouse strains





Substitution of M for H in the RHZ regimen in mice – data from 3 institutions, using low- & high-dose infection models

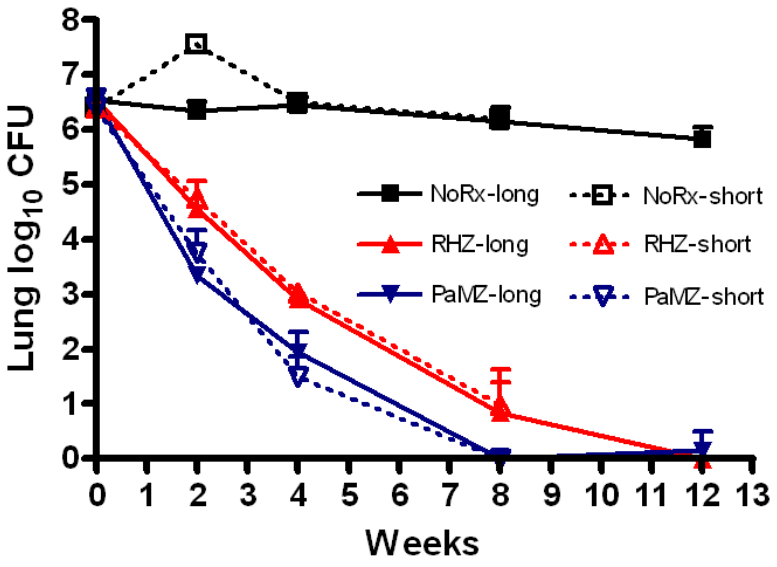


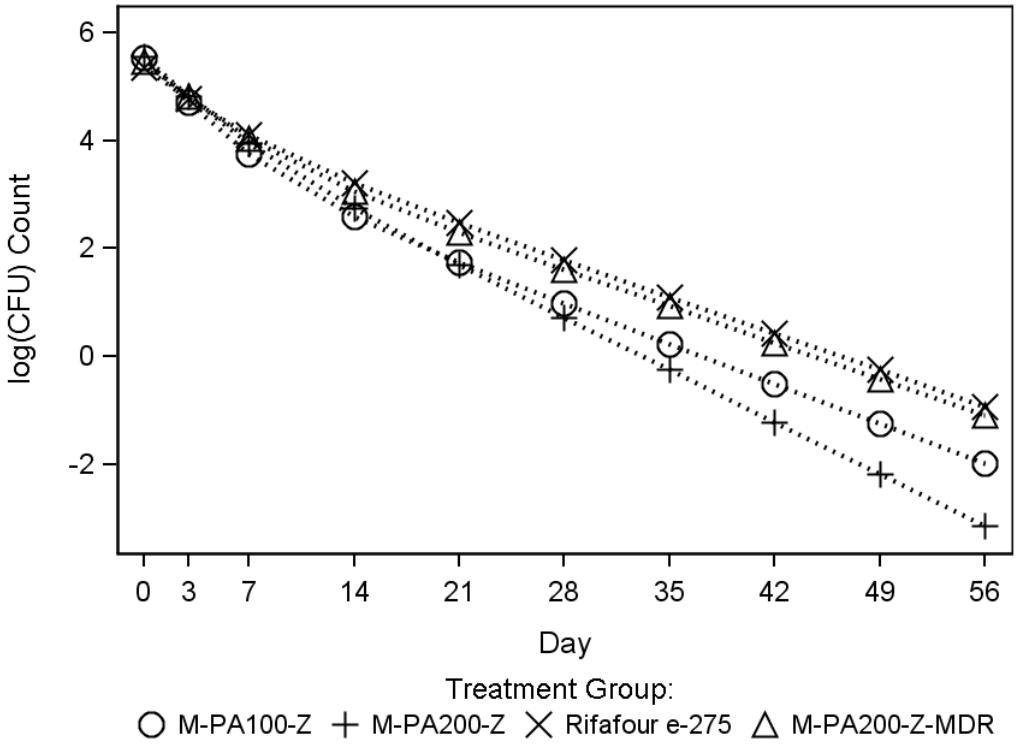
Correspondence between results of REMox-TB trial and those in mice

- In REMox-TB, substitution of M for H or E resulted in faster sputum conversion but did not permit shortening the duration of treatment to 4 months

- In mice, substitution of M for H or E reduced CFU cts more rapidly, but relapse rates were inevitably higher when RMZ(E) or RHZM duration was reduced by 2 months relative to RHZ(E)
- Results in C3HeB/FeJ mice may be closer to those of REMox-TB
  - smaller difference for RMZE relative to RHZE
  - no difference between RHZM and RMZE
- The most severely affected C3HeB/FeJ mice may best represent pts most likely to relapse

Bacterial killing with $Pa_{50}MZ$ vs. RHZ: mouse vs. NC-002 results

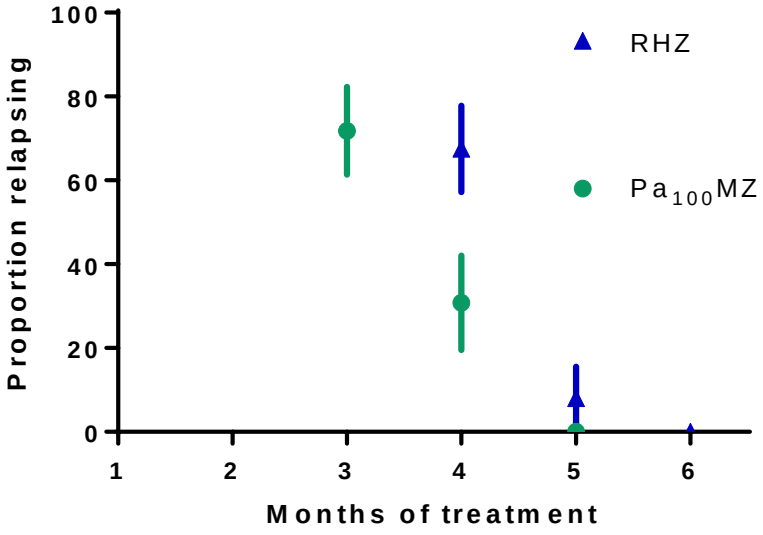




Long = 6 wks from low-dose infxn to treatment onset
Short = 2 wks from high-dose infxn to treatment onset

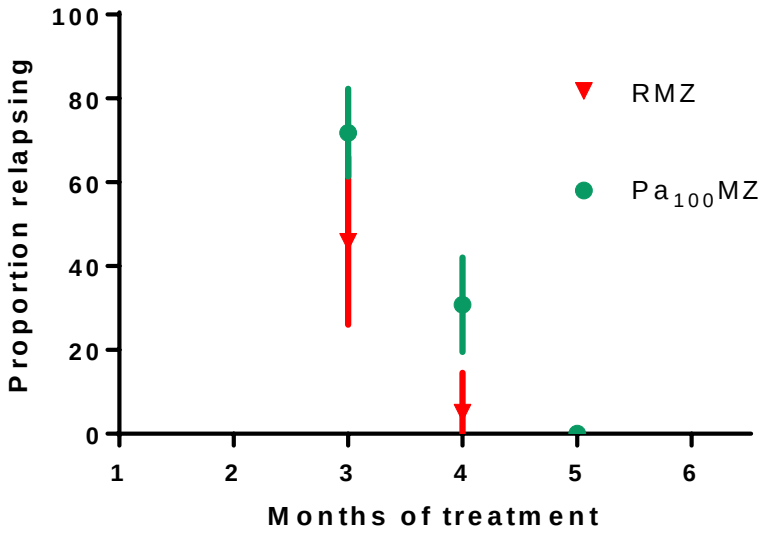
PaMZ vs. RHZ, RMZ
HDA model in BALB/c mice – relapse data

PaMZ vs. RHZ



Data from 5 experiments (3 with Pa 100mpk, 2 with Pa 50mpk)

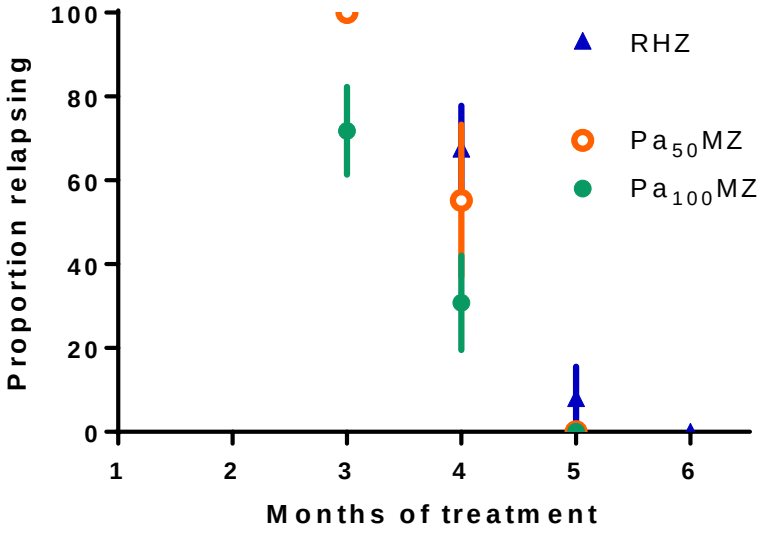
PaMZ vs. RMZ



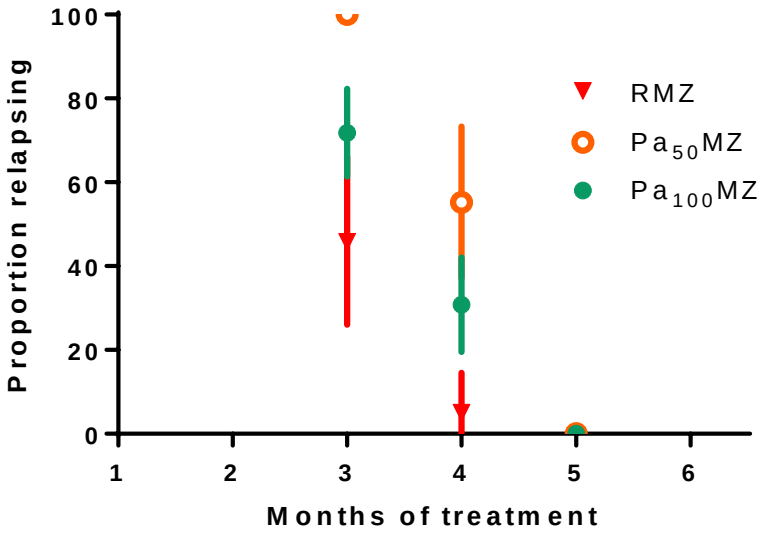
Data from 5 experiments (2 with head-to-head data)

PaMZ vs. RHZ, RMZ
HDA model in BALB/c mice – relapse data

PaMZ vs. RHZ



PaMZ vs. RMZ



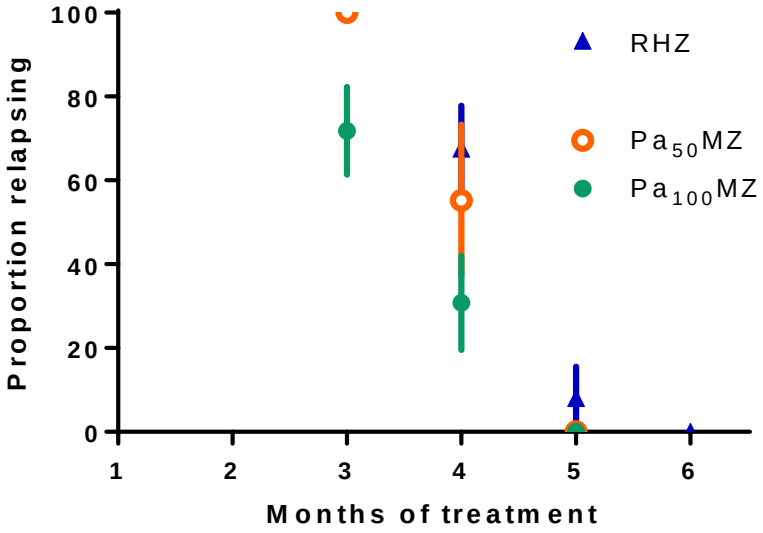
The treatment shortening effect is between 0-1 month when compared to RHZ and depends on the Pa dose.
PaMZ is no more effective than RMZ and appears less effective than RMZ when the Pa dose is 50 mpk.

Data from 5 experiments (3 with Pa 100mpk, 2 with Pa 50mpk)

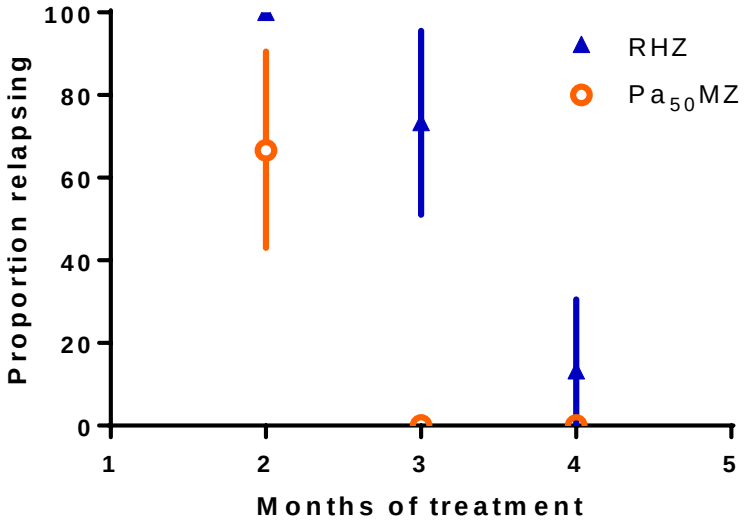
Data from 5 experiments (2 with head-to-head data)

PaMZ vs. RHZ
HDA model in BALB/c mice – relapse data

Standard model



Lower dose infxn

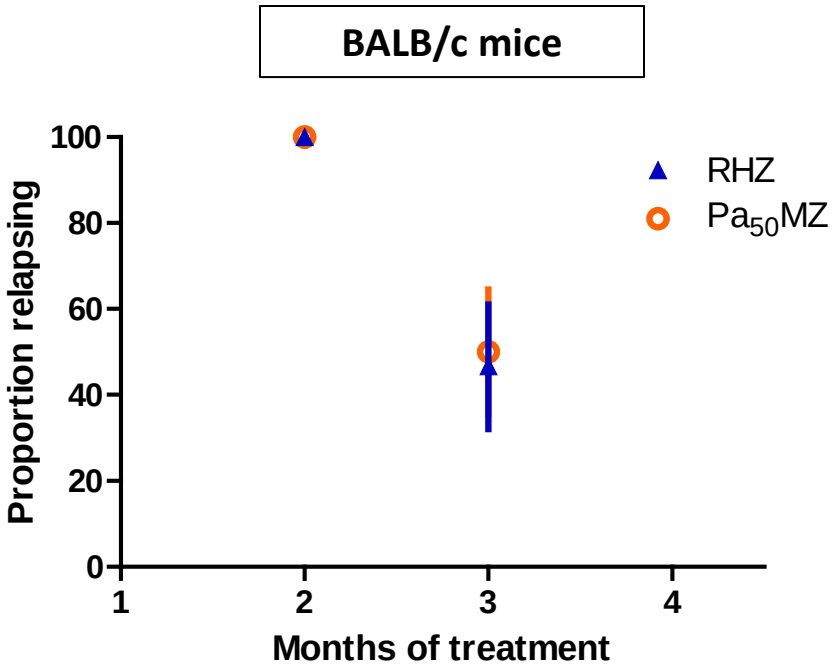


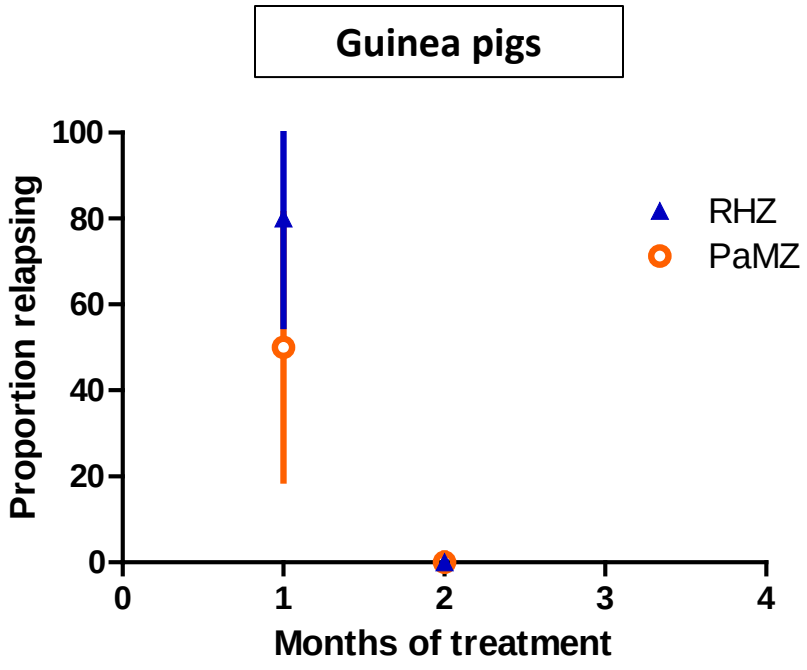
In one experiment in which a lower-dose aerosol infection was used in a 14-day incubation model to match Day 0 CFU counts with a low-dose chronic (42-day) infection model (right panel), the treatment shortening effect of PaMZ was at least 1 month.

Data from 5 experiments (3 with Pa 100mpk, 2 with Pa 50mpk)

Data from 1 experiment

PaMZ vs. RHZ
Chronic LDA model in BALB/c mice, guinea pigs





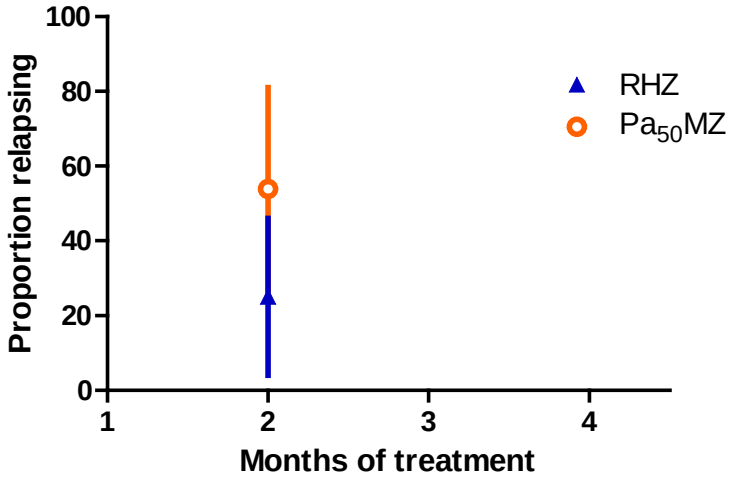
In the chronic low-dose aerosol model in BALB/c mice, $Pa_{50}MZ$ is equivalent to RHZ.
In the chronic guinea pig model, the treatment shortening effect of PaMZ is between 0 and 1 month.

Data from 3 experiments

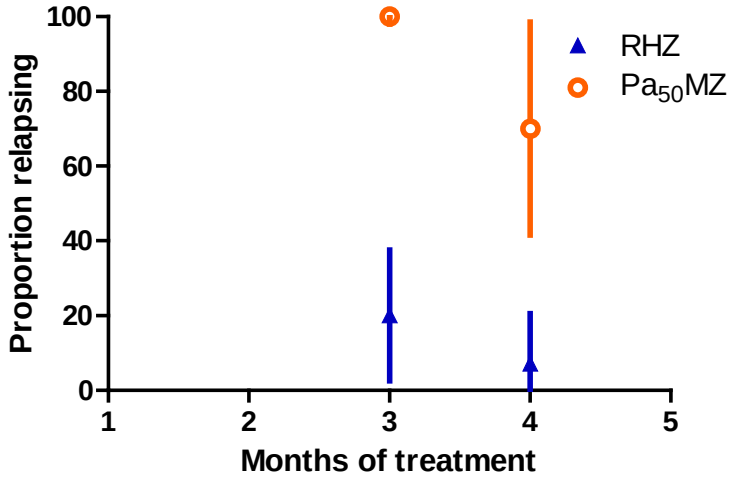
Dutta et al, AAC (2013); 57:3910

PaMZ vs. RHZ
LDA model in C3HeB/FeJ mice – relapse data

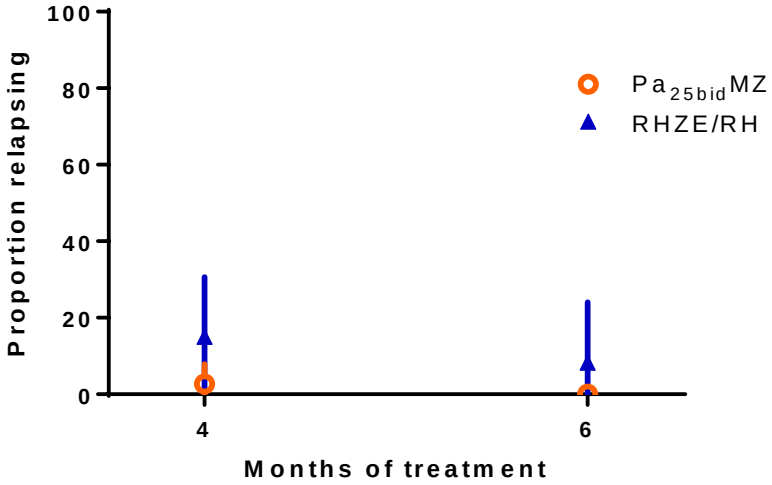
Lower dose infection



Higher dose infxn



Higher dose infxn

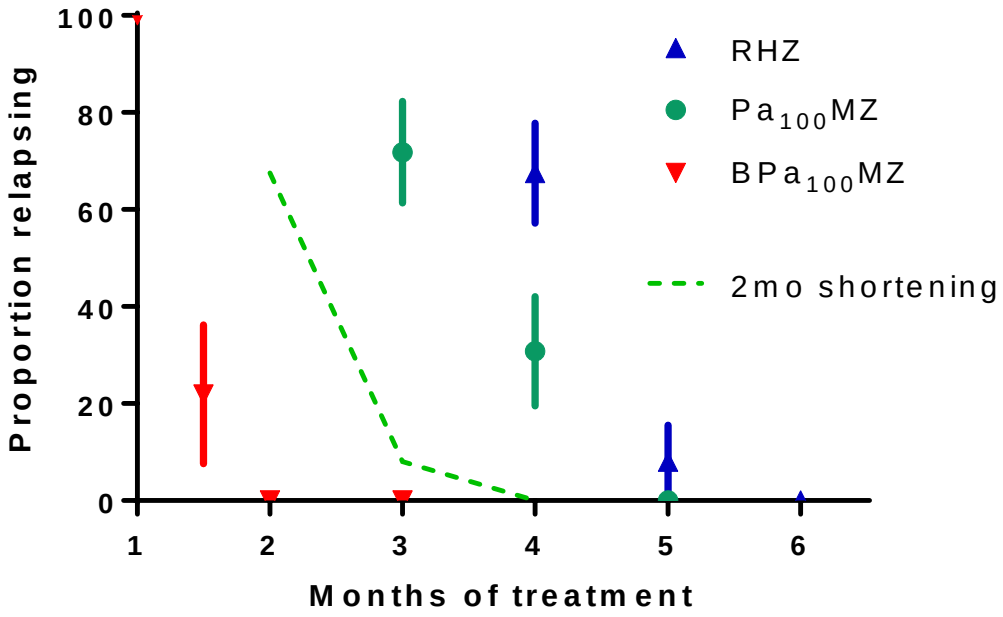


In C3HeB/FeJ mice, $Pa_{50}MZ$ is less effective than RHZ except in one expt in which Pa, M and Z dosing was divided twice daily.

Conclusions re: PaMZ

- Results of the abbreviated STAND trial comparing 2RHZE/4RH to 4PaMZ and 6PaMZ should be available soon & will provide a basis for comparison of mouse and human results
- In our standard high-dose aerosol model in BALB/c mice:
  - PaMZ requires 0-1 month less treatment to cure compared to RHZ, and this effect is somewhat dose-dependent
  - PaMZ requires 0-1 month **more** treatment to cure compared to RMZ

BPaMZ vs. PaMZ vs. RHZ
HDA model in BALB/c mice

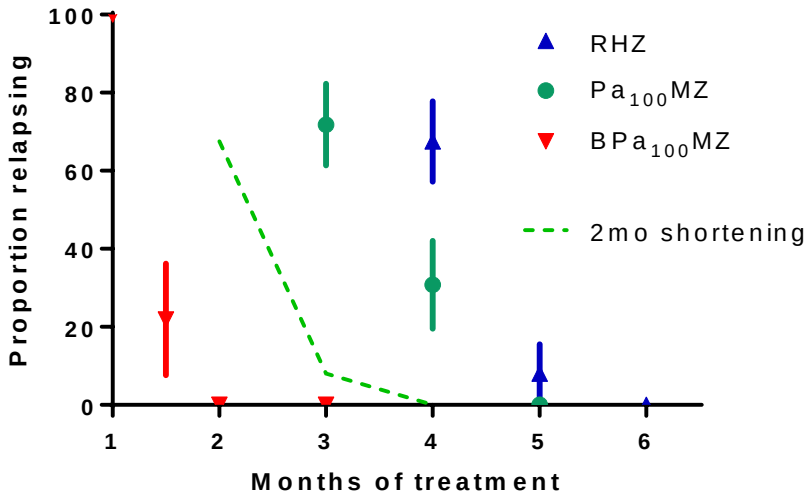


In the high-dose aerosol model in BALB/c mice, $BPa_{100}MZ$ shortens the duration of treatment by 3-3.5 months compared to RHZ.

BPaMZ data from 2 experiments

BPaMZ vs. PaMZ vs. RHZ
HDA model in BALB/c mice

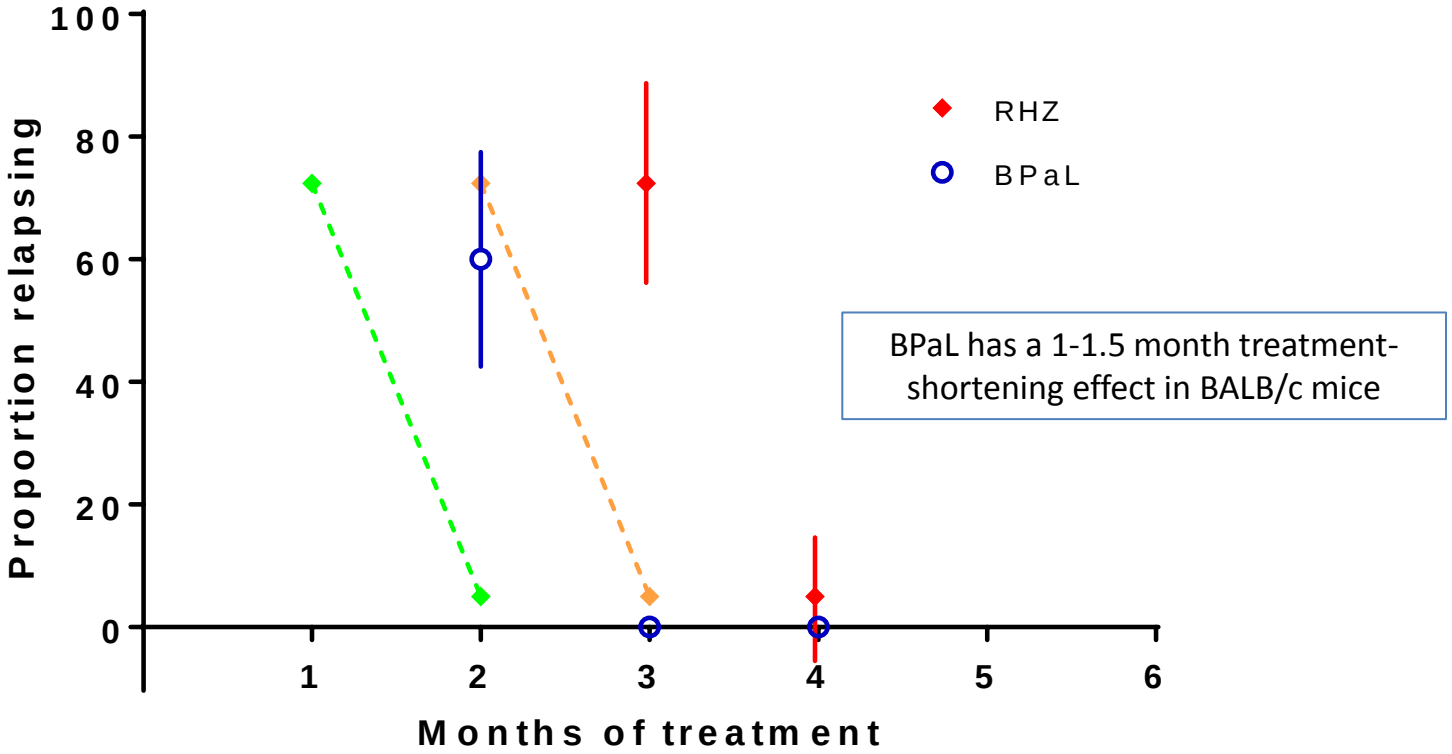
All expts evaluating PaMZ and/or BPaMZ



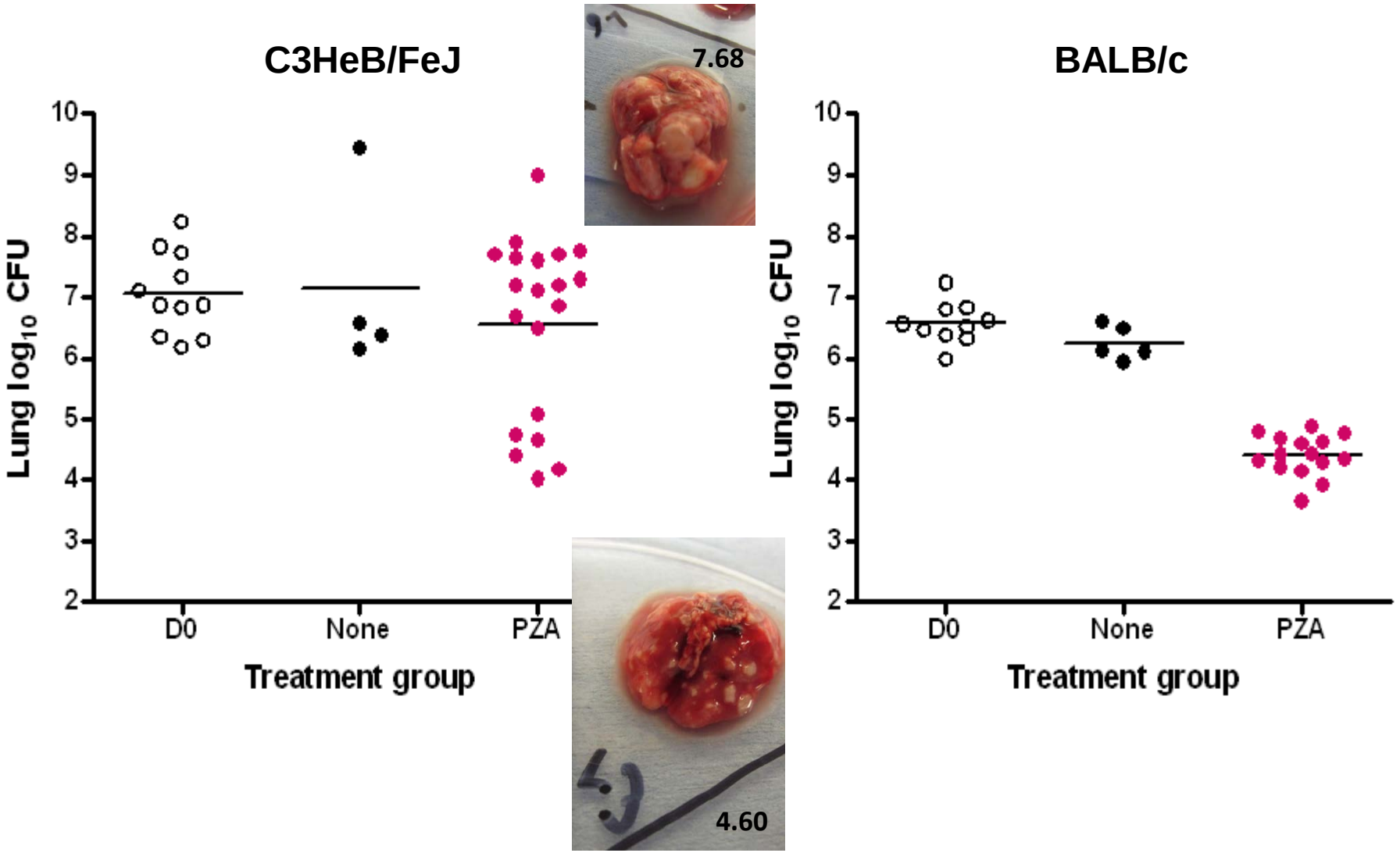
In the high-dose aerosol model in BALB/c mice, $BPa_{100}MZ$ shortens the duration of treatment by 3-3.5 months compared to RHZ.

BPaMZ data from 2 experiments

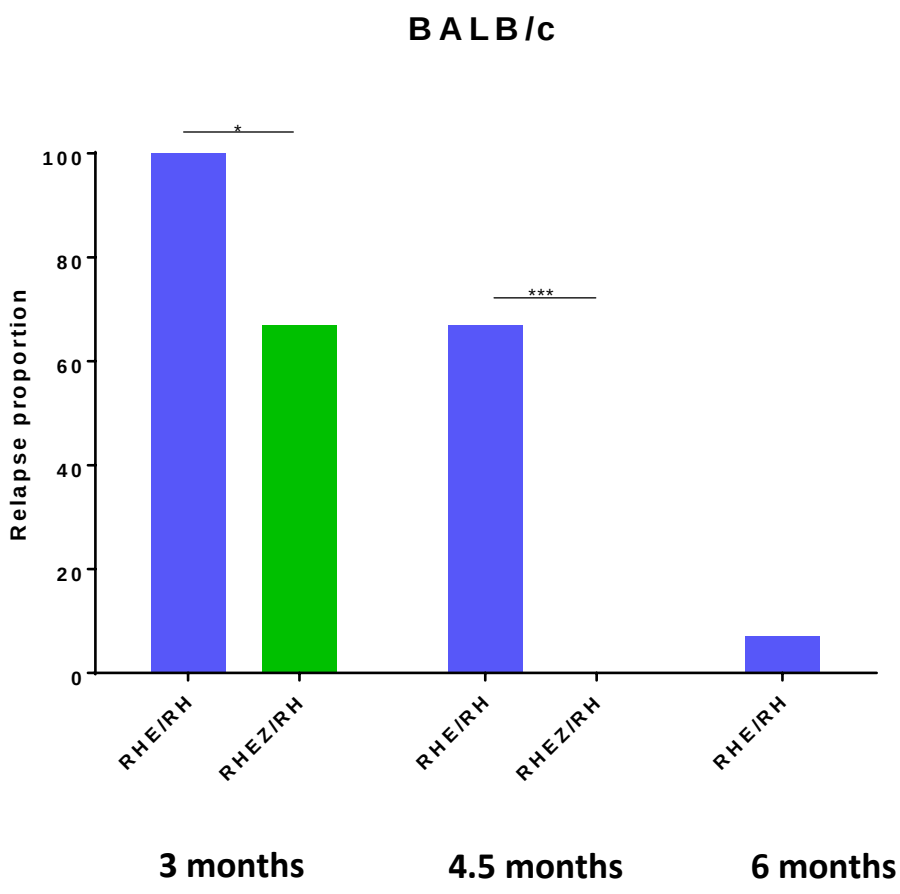
BPaL vs. RHZ
HDA model in BALB/c mice

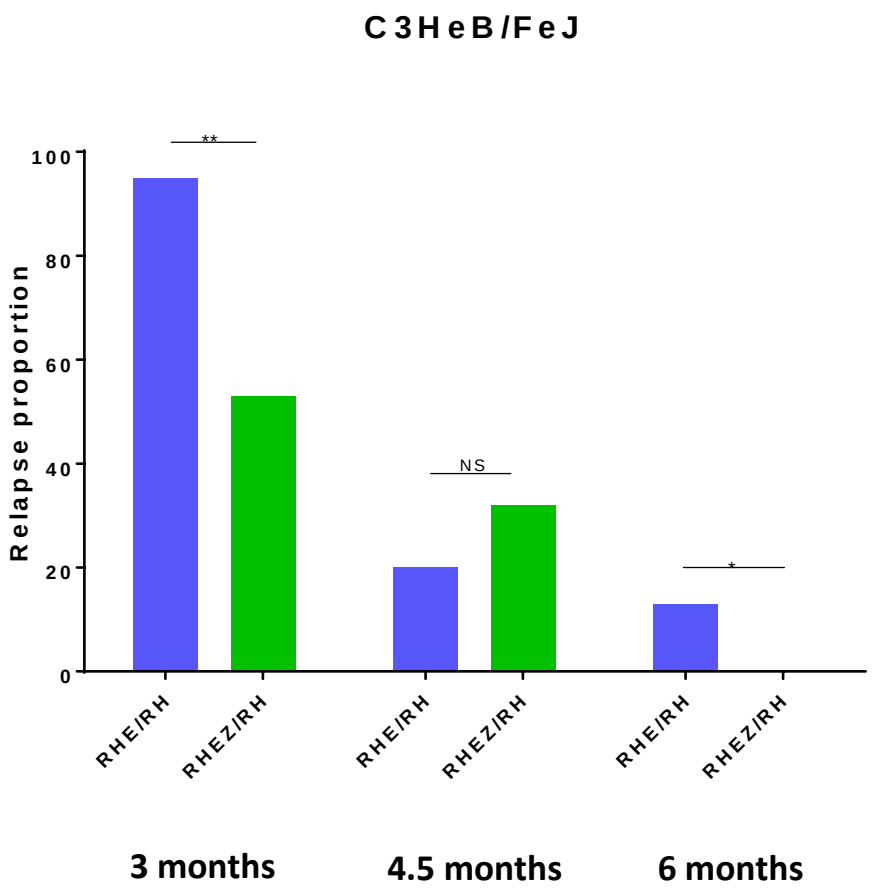


“Dichotomous” activity of PZA in C3HeB/FeJ mice

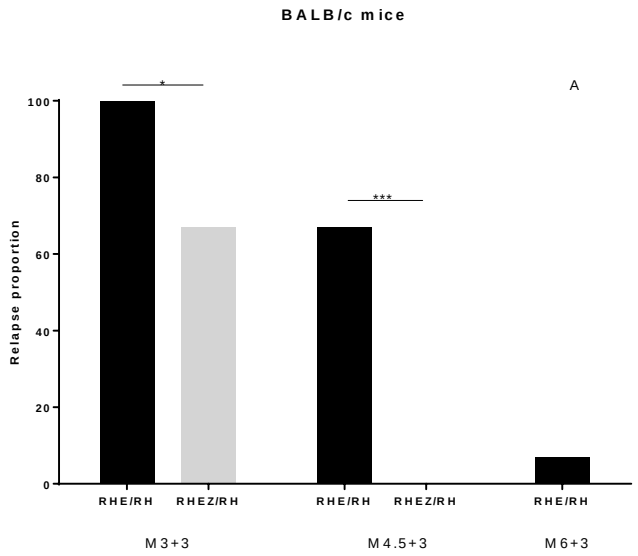


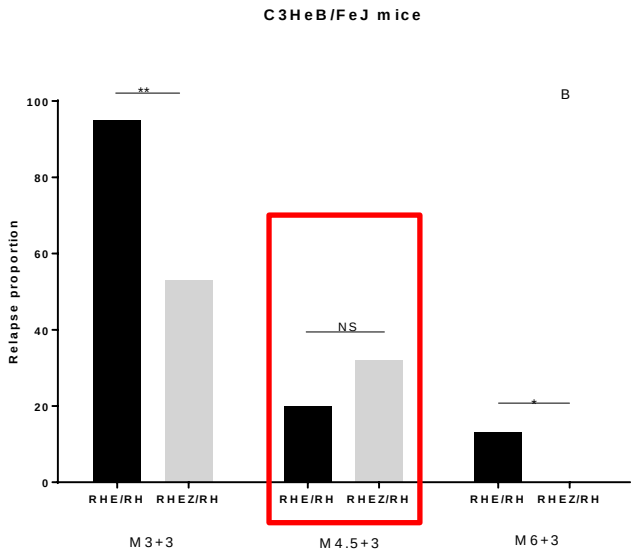
Z adds sterilizing activity to RHE in BALB/c and C3HeB/FeJ mice

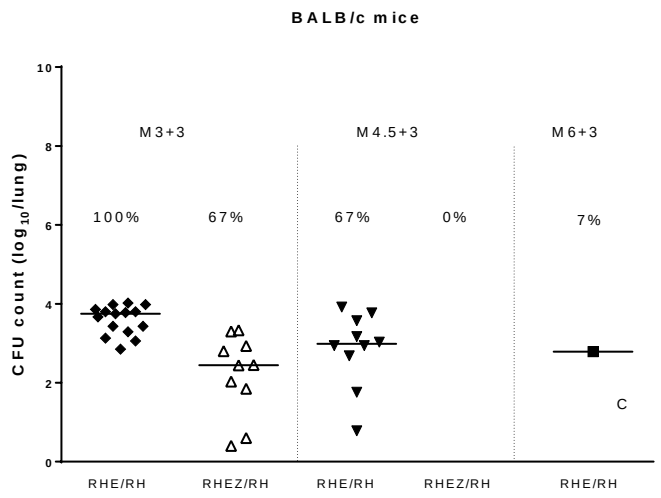


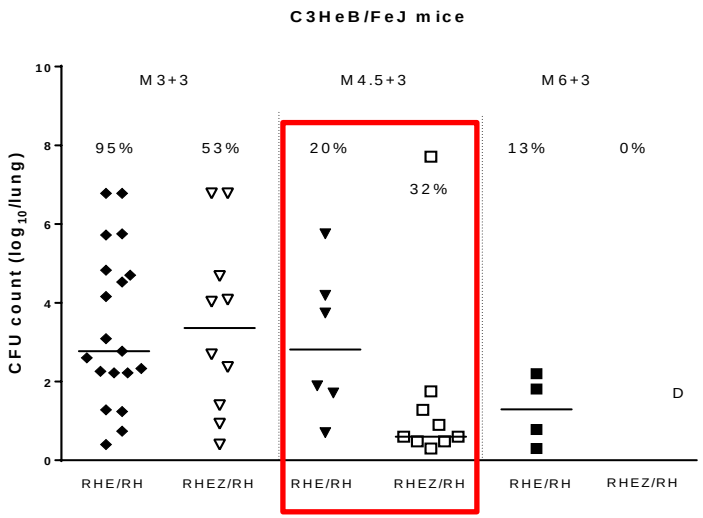


Sterilizing activity of Z in 1st-line regimen in 2 mouse strains

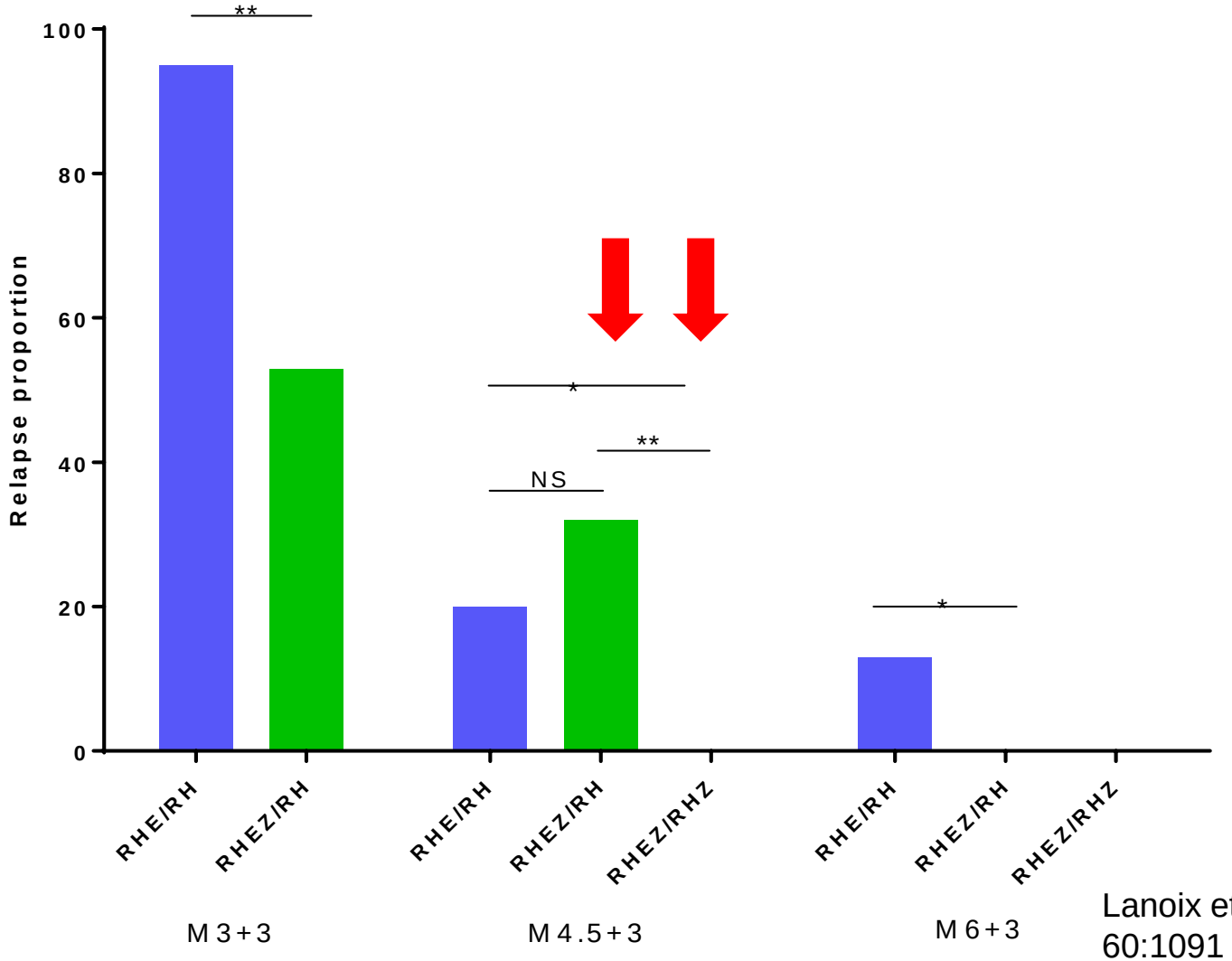








Increasing duration of PZA increases sterilizing effect in C3HeB/FeJ mice



Lanoix et al, AAC (2016); 60:1091

Z Adds Sterilizing Activity to RHE in BALB/c and C3HeB/FeJ Mice

	Does use of Z for 2 months reduce relapses compared to no Z?			Does extending Z beyond 2 months reduce relapses?
	3 mo.	4.5 mo.	6 mo.	3-4.5 mo.
BALB/c	Yes	Yes	Yes	No
Kramnik	Yes	No*	Yes	Yes†

*Z was associated with lower mean CFU count among relapsing mice
†2RHEZ/2.5RHZ was associated with fewer relapses than 2RHEZ/2.5RH

Lanoix et al, AAC (2016); 60:1091

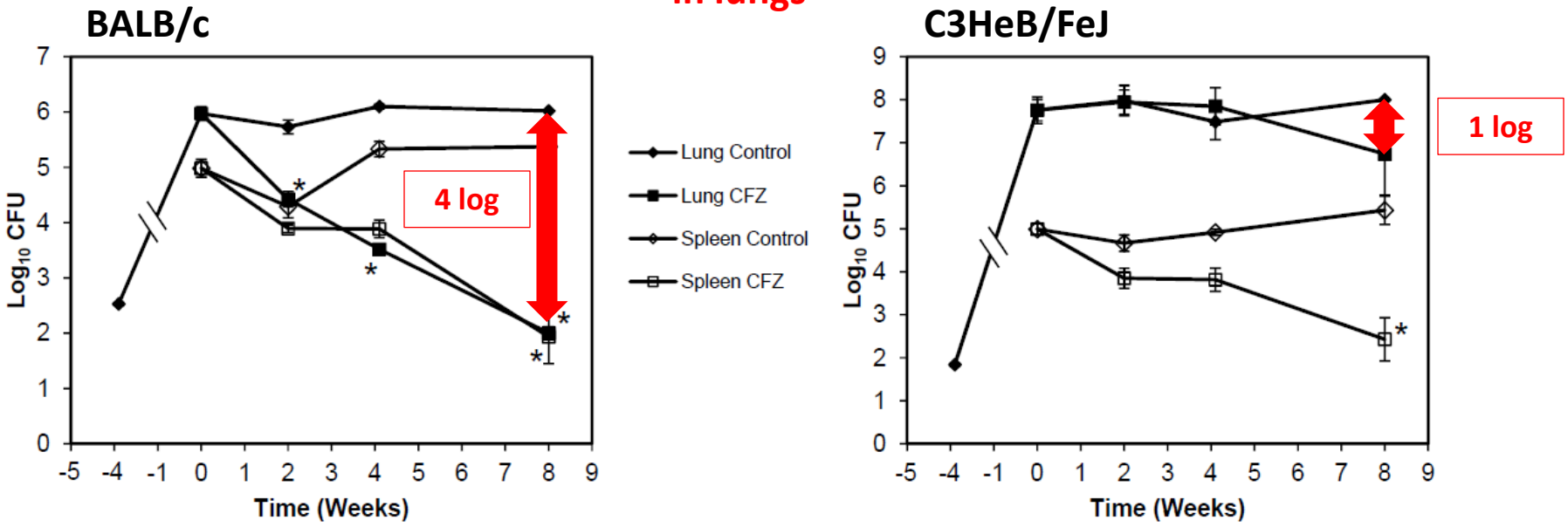
Study	*Regimen**	*Follow-up* (months)	*Patients assessed*	*Bacterio-logical relapses*
East African/BMRC (current)	2SHRZ/HRZ	24	40	0
	2SHRZ/HR	24	40	1 (2%)
Singapore/BMRC (1981)	2SHRZ/HRZ	24	78	0
	2SHRZ/HR		80	2 (2%)

Can we really conclude that continuing Z beyond 2 months would not benefit the most difficult-to-cure patients?

Fox, Br J Dis Chest (1981); 75:331

Activity of clofazimine in C3HeB/FeJ mice

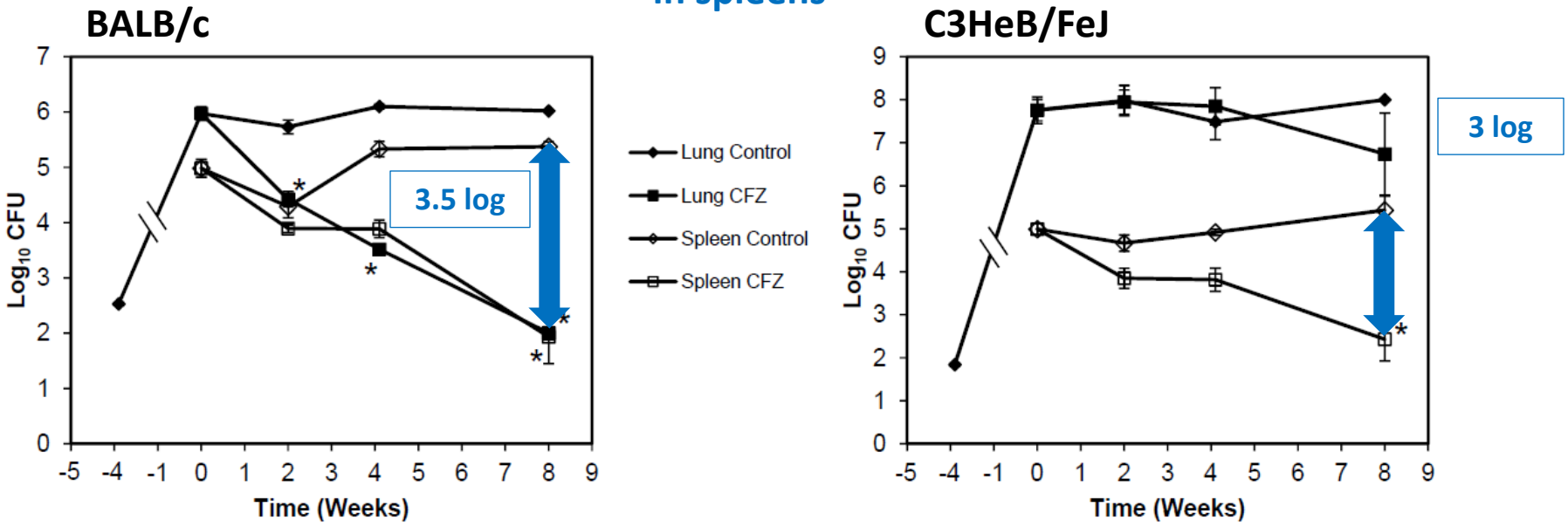
CFZ effect size in lungs



SM Irwin et al, AAC 2014; 58:4026

Activity of clofazimine in C3HeB/FeJ mice

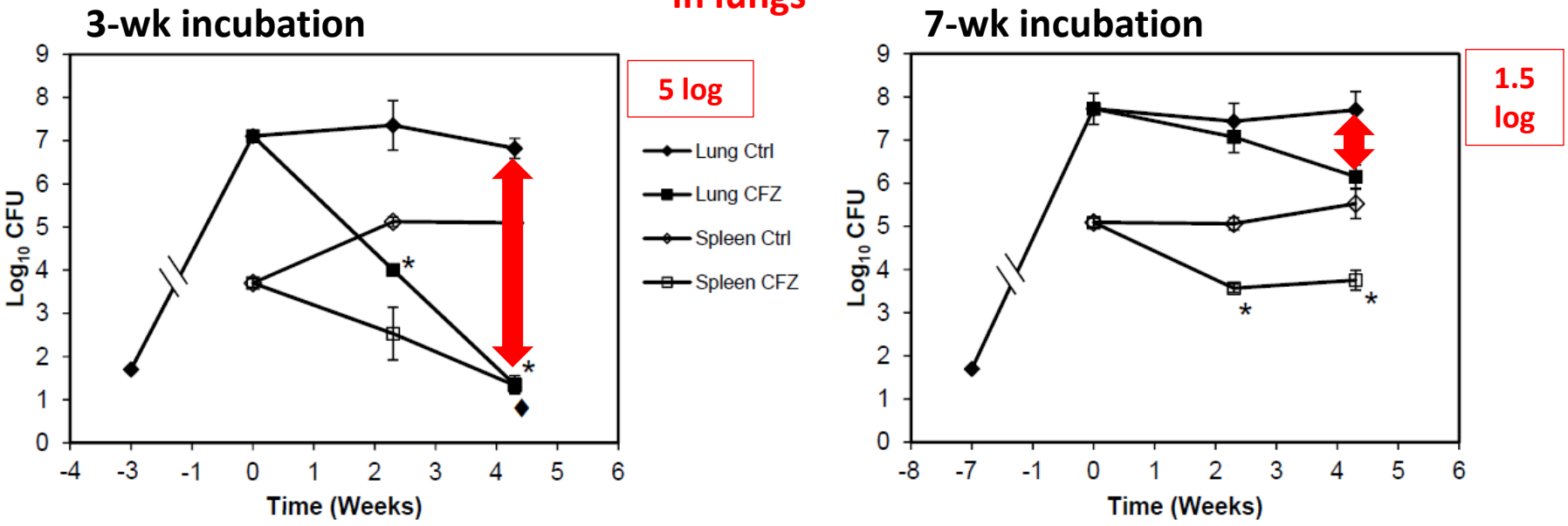
CFZ effect size in spleens



SM Irwin et al, AAC 2014; 58:4026

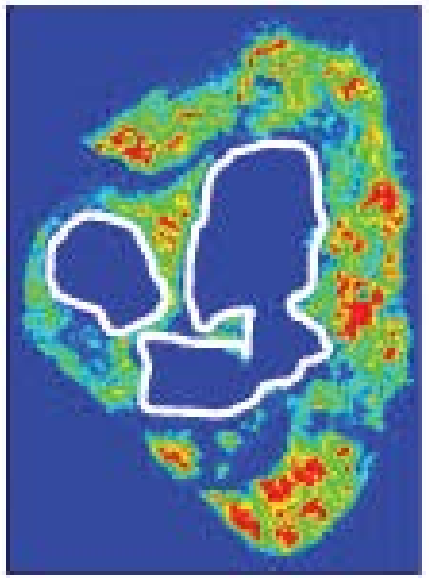
Activity of clofazimine in C3HeB/FeJ mice

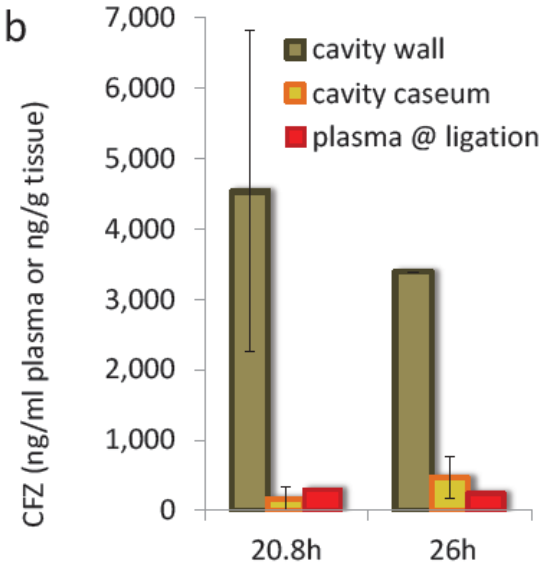
CFZ effect size in lungs



Compartmentalized activity of CFZ – PK/distribution

- Slow, steady accumulation in adipose tissue & macrophages
  - pH-dependent ion trapping in lysosomes
- Poor distribution into caseum relative to cavity wall





Data inventory

- Focus first on mouse strains other than C3HeB/FeJ ("Kramnik")
- Inventory identified a variety of relapse-based pre-clinical studies with corresponding clinical trial outcomes data

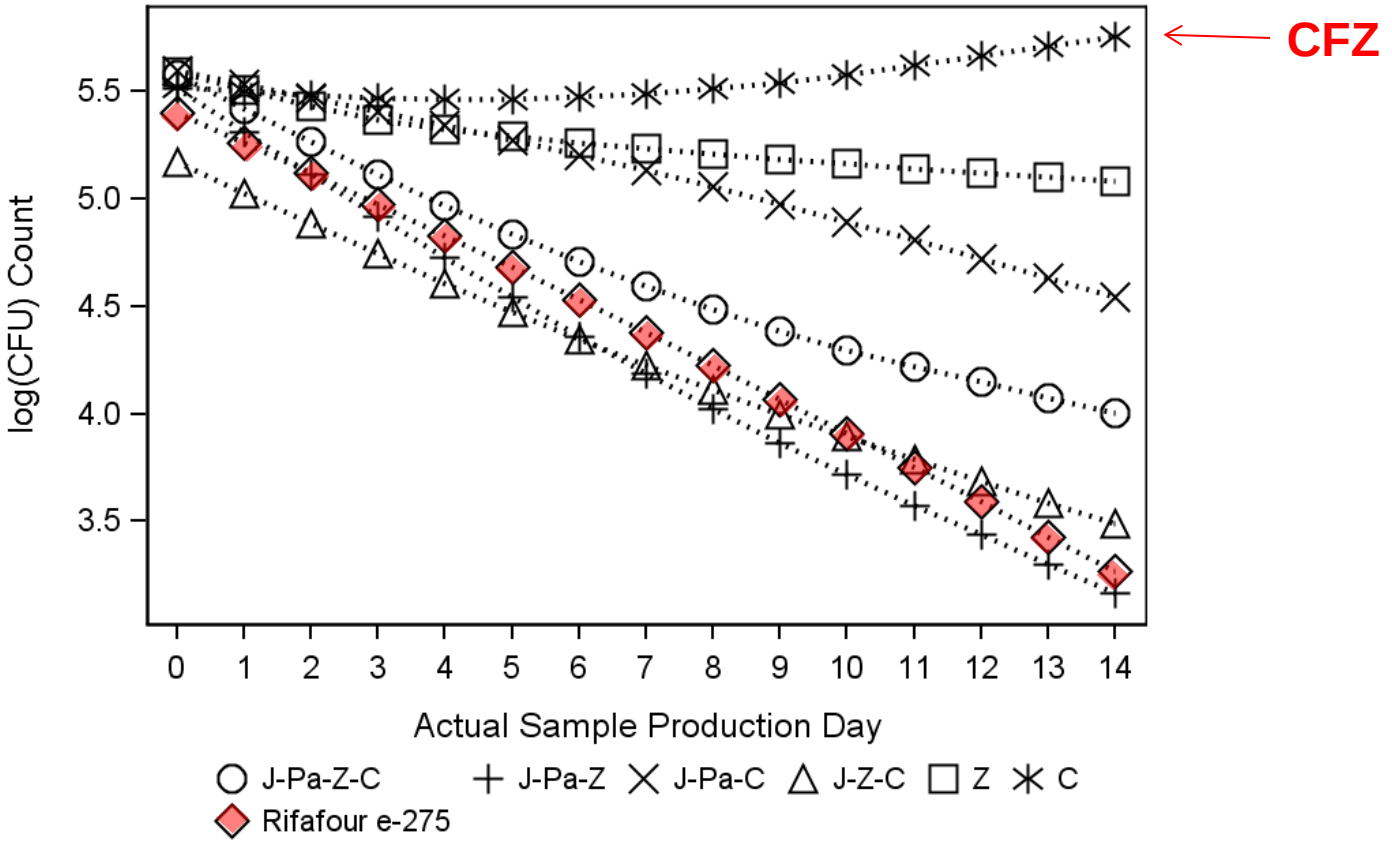
Test regimen intervention	Regimen comparison	# of expts
Combining INH+STR	HS vs. H or S monotherapy	1
Shortening duration of INH+STR	6HS vs. 18HS	1
Adding RIF to INH+STR or INH+EMB+PZA	HR (or HRS or HREZ) vs. HS (or HEZ)	4
Adding STR to INH+RIF	HRS vs. HR	1
Adding PZA to INH+RIF (±STR/EMB)	HRZ (or HRSZ or HREZ) vs. HR (or HRS or HRE)	4
Shortening duration of PZA	2HREZ/4RH vs. 6HREZ	1
Increasing dose of RIF	High-dose R plus HEZ vs. HREZ	2
Extending dosing interval of 1st-line Rx	HREZ (2/7) vs. HREZ (daily)	1
Replacing EMB with MXF	HRMZ vs. HRZ(E)	3
Replacing INH with MXF	MRZ(E) vs. HRZ(E)	10
Replacing RIF with RPT	HPZ(E) vs. HRZ(E)	7
Replacing RIF+EMB with RPT+MXF	HPMZ vs. HRZ	3
Replacing RIF with RPT and extending dosing interval (in continuation phase)	HP(1/7) cont phase vs. HR(2/7)	2
Comparing INH+RIF+PZA+EMB with PMD+MXF+PZA	PaMZ vs. HRZ(E)	8

Summary points

- An initial step to address the “translational gap” is to learn what data from what models analyzed in what way best inform key trial design decisions.
- Evidence-based validation of pre-clinical models is important:
  - to confidently place preclinical models on the critical development path,
  - to increase the efficiency of regulatory interactions,
  - to set a precedent for objective, data-driven processes to apply to other models (e.g., C3HeB/FeJ mouse, marmoset), and
  - to identify gaps in knowledge & in existing tools to drive future research.
- Evaluation of sterilizing mouse models is the appropriate first step for *in vivo* models, with other models to follow

Clofazimine has no EBA in TB patients

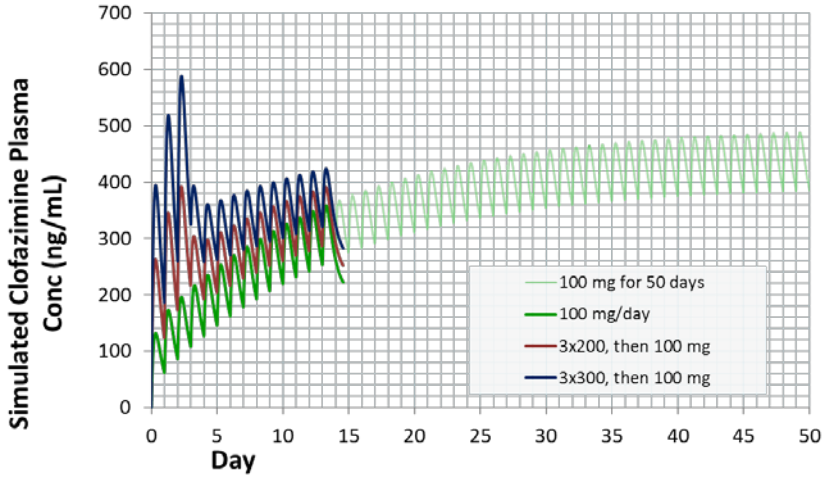
Serial sputum colony counts over 1st 14 days of treatment



A Diacon et al, AJRCCM 2015

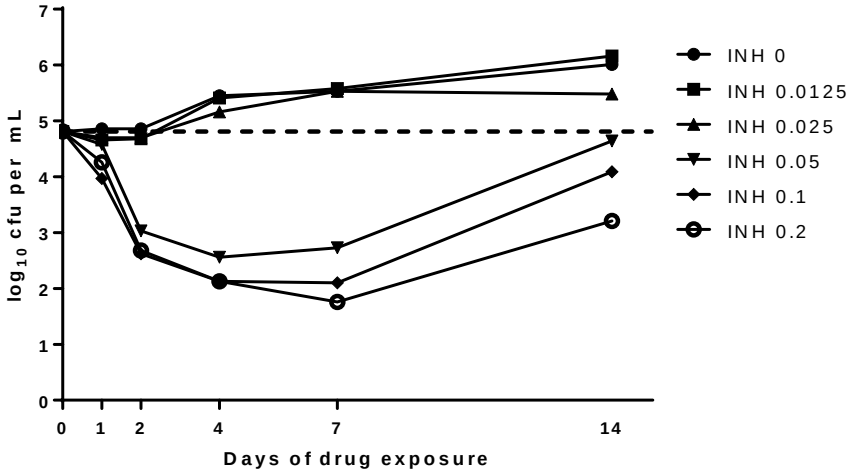
Compartmentalized activity of CFZ and slow onset of effect – PK/distribution

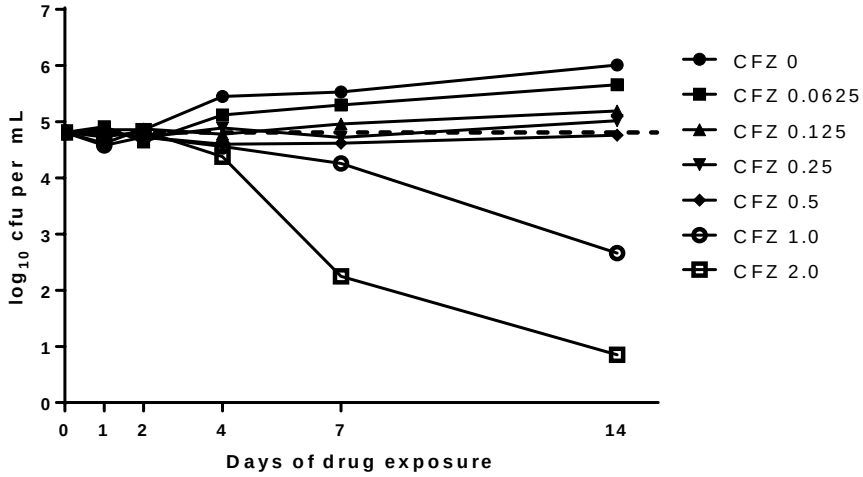
- >1 month to reach steady state
- Slow, steady accumulation in adipose tissue & macrophages
  - pH-dependent ion trapping in lysosomes



D Everitt et al, derived from Schaad-Lanyi et al

In vitro EBA of INH and CFZ at similar multiples of their MICs

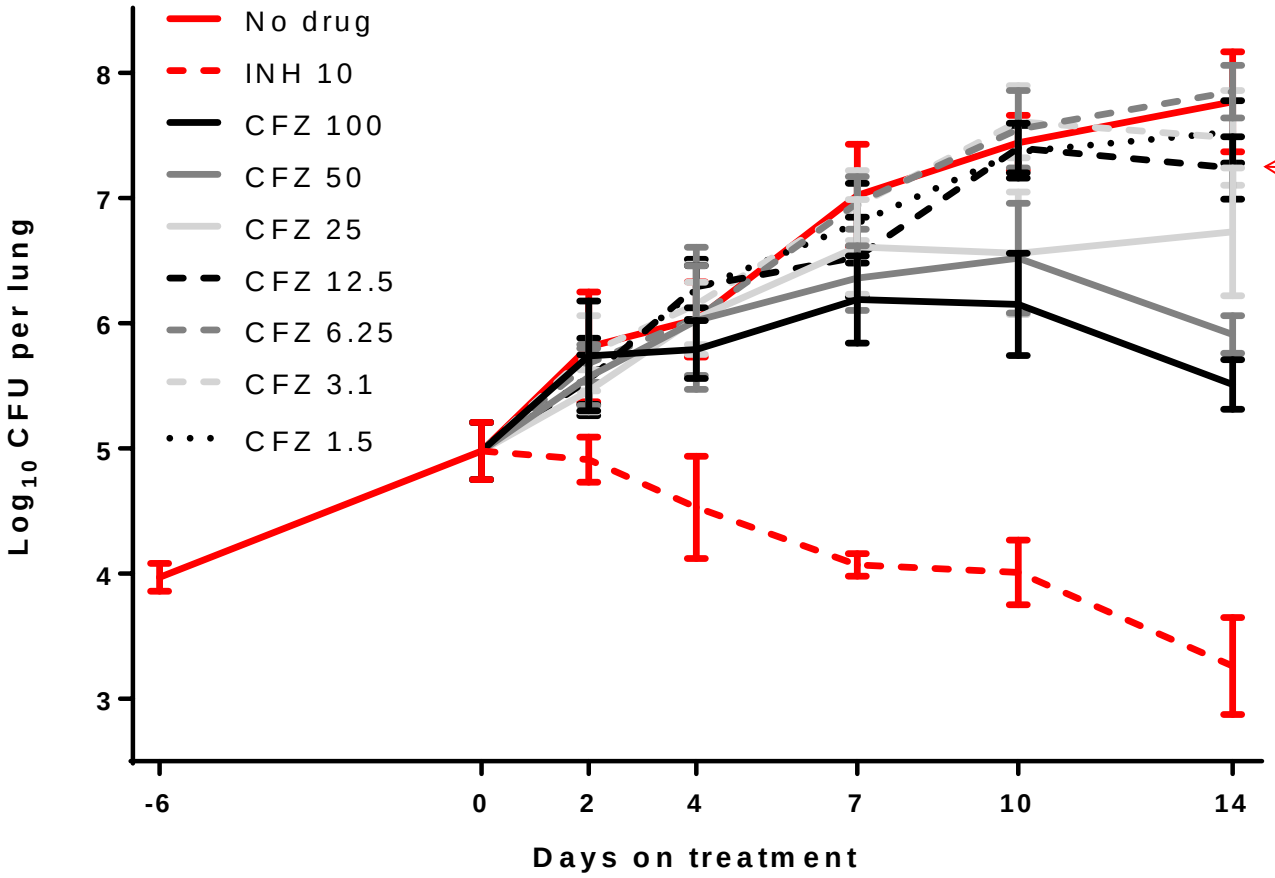




- MICs: INH = 0.05 µg/mL, CFZ = 0.25 µg/mL

N. Ammerman et al, accepted for publication

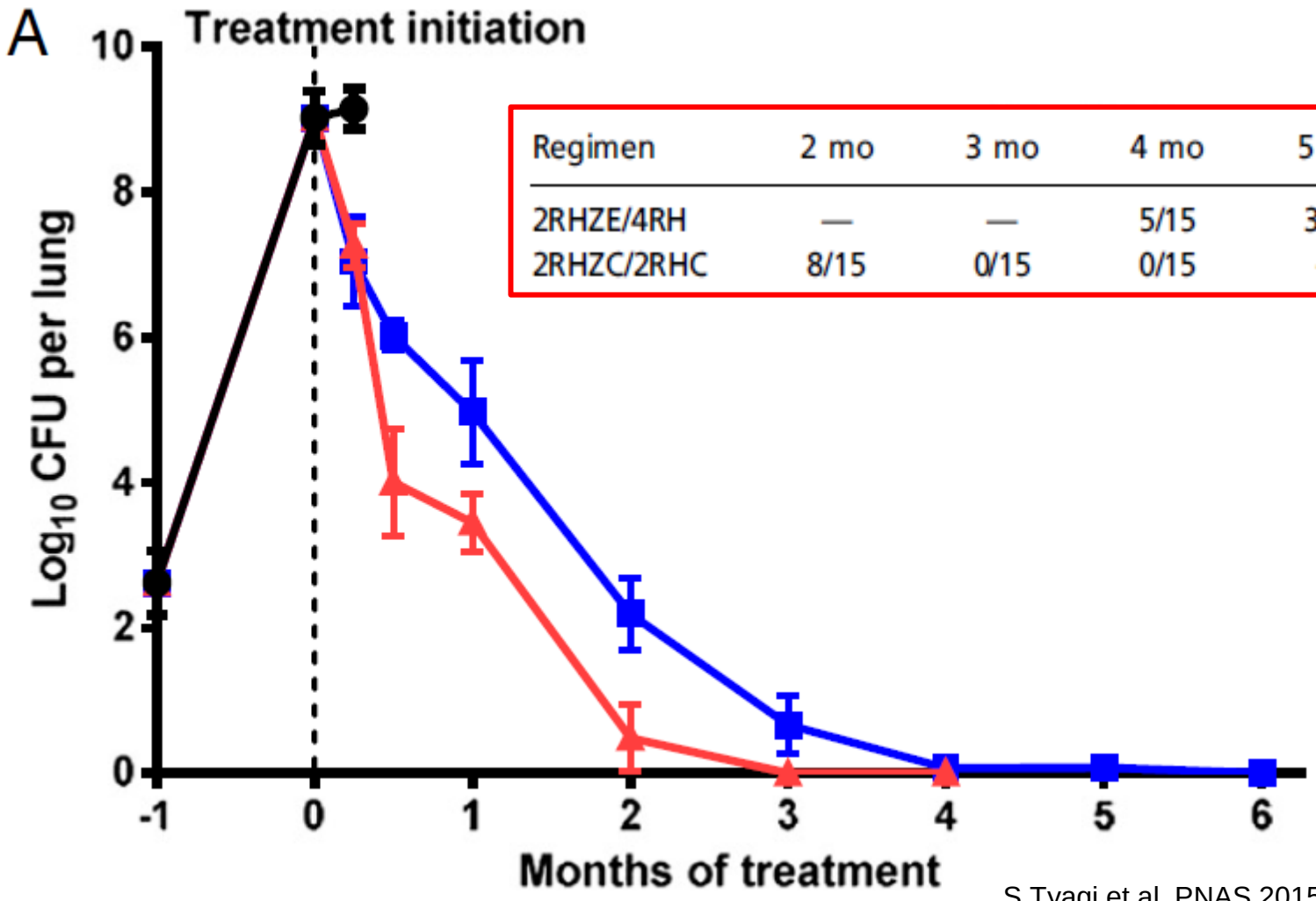
EBA of INH and CFZ in BALB/c mice



100 mg human dose equivalent

N. Ammerman et al, accepted for publication

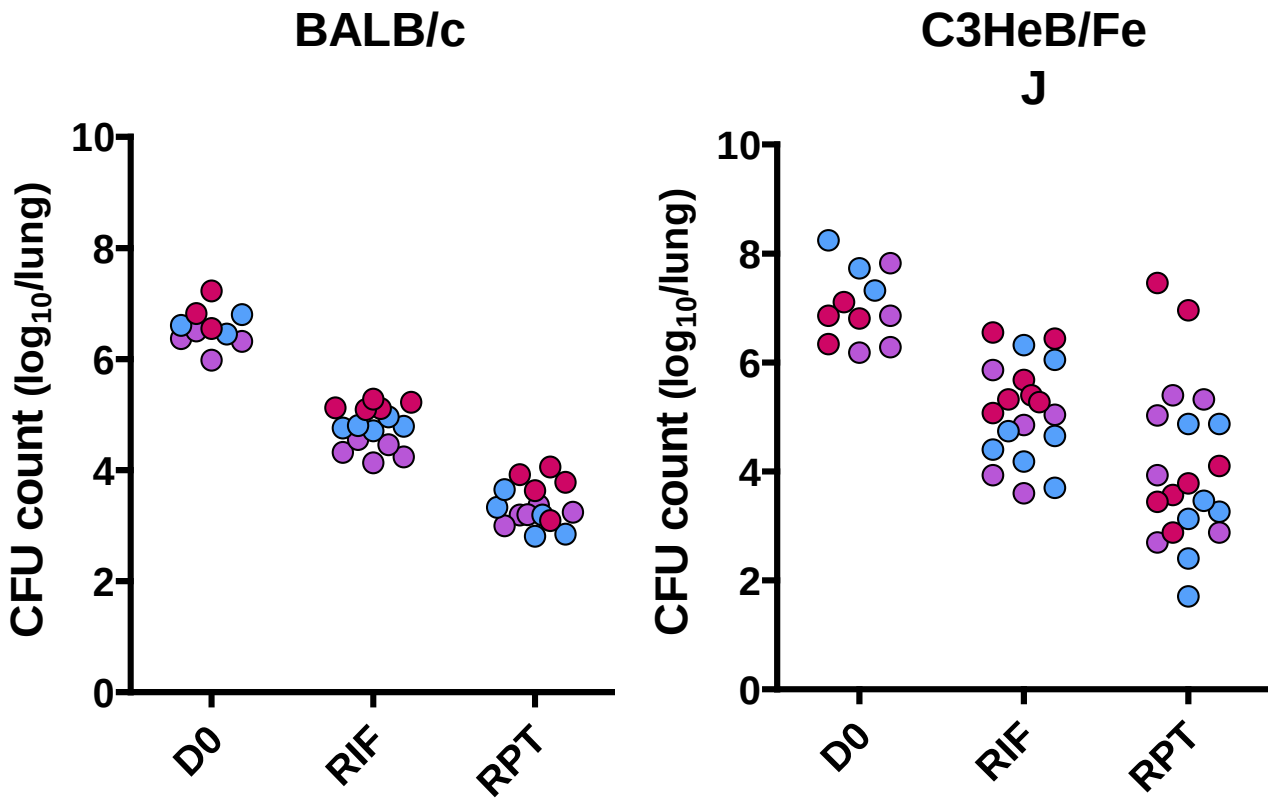
Incorporation of CFZ into the 1st-line regimen in mice



Regimen	2 mo	3 mo	4 mo	5 mo	6 mo
2RHZE/4RH	—	—	5/15	3/15	1/10
2RHZC/2RHC	8/15	0/15	0/15	—	—

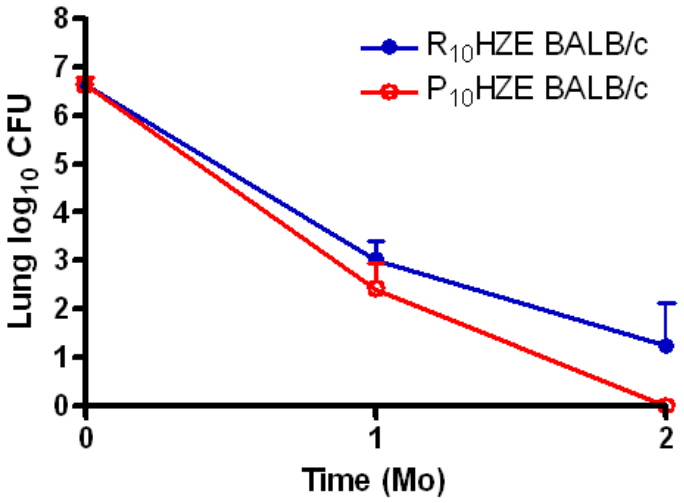
S Tyagi et al, PNAS 2015; 112:869

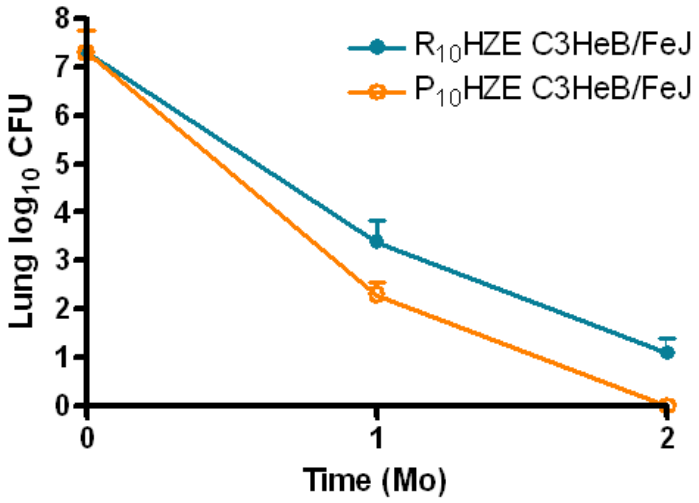
Comparative activity of RIF and RPT in BALB/c and C3HeB/FeJ mice over 4 wks of treatment



RHZE vs. PHZE in 2 mouse strains

CFU counts





Relapse

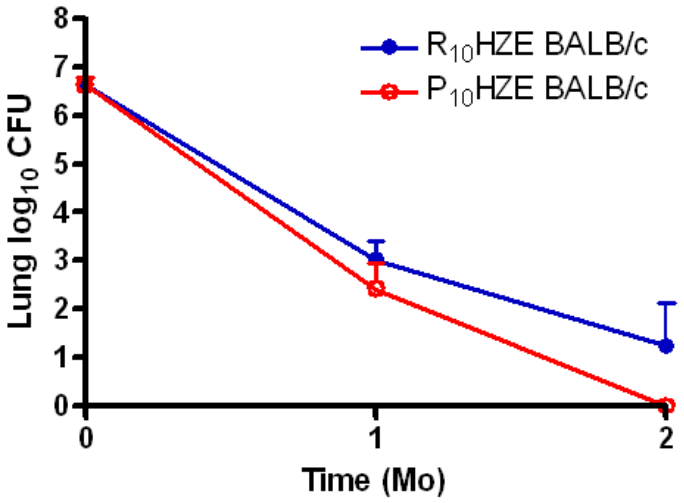
Drug regimen	Mouse strain	% (proportion) of mice with relapsing after treatment for:			
		1 month	2 months	3 months	4 months
R_{10}HZE	BALB/c	ND	100% (15/15)	47% (7/15)	13% (2/15)
	C3HeB/FeJ	ND	100% (15/15)	86% (12/14)	7% (1/15)
P_{10}HZE	BALB/c	100% (14/14)	7% (1/15)	0% (0/15)	
	C3HeB/FeJ	100% (13/13)	21% (3/14)	33% (5/15)	

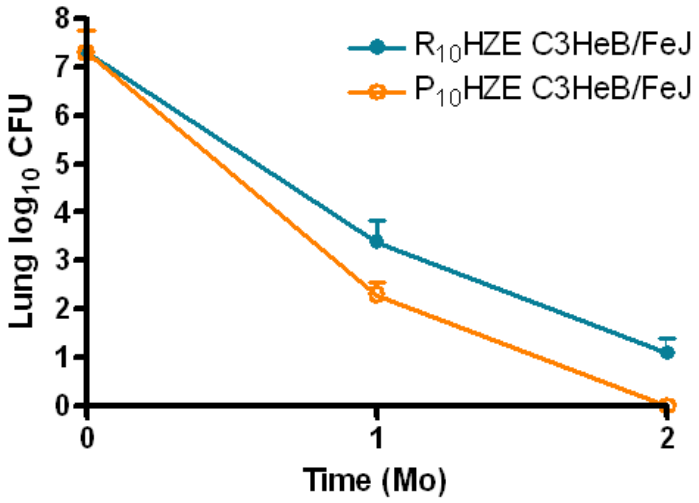
2 month shortening with P in BALB/c

Rosenthal et al, AAC 2012; 56:4331

RHZE vs. PHZE in 2 mouse strains

CFU counts





Relapse

Drug regimen	Mouse strain	% (proportion) of mice with relapsing after treatment for:			
		1 month	2 months	3 months	4 months
R_{10}HZE	BALB/c	ND	100% (15/15)	47% (7/15)	13% (2/15)
	C3HeB/FeJ	ND	100% (15/15)	86% (12/14)	7% (1/15)
P_{10}HZE	BALB/c	100% (14/14)	7% (1/15)	0% (0/15)	
	C3HeB/FeJ	100% (13/13)	21% (3/14)	33% (5/15)	

< 1 month shortening with P in C3HeB/FeJ

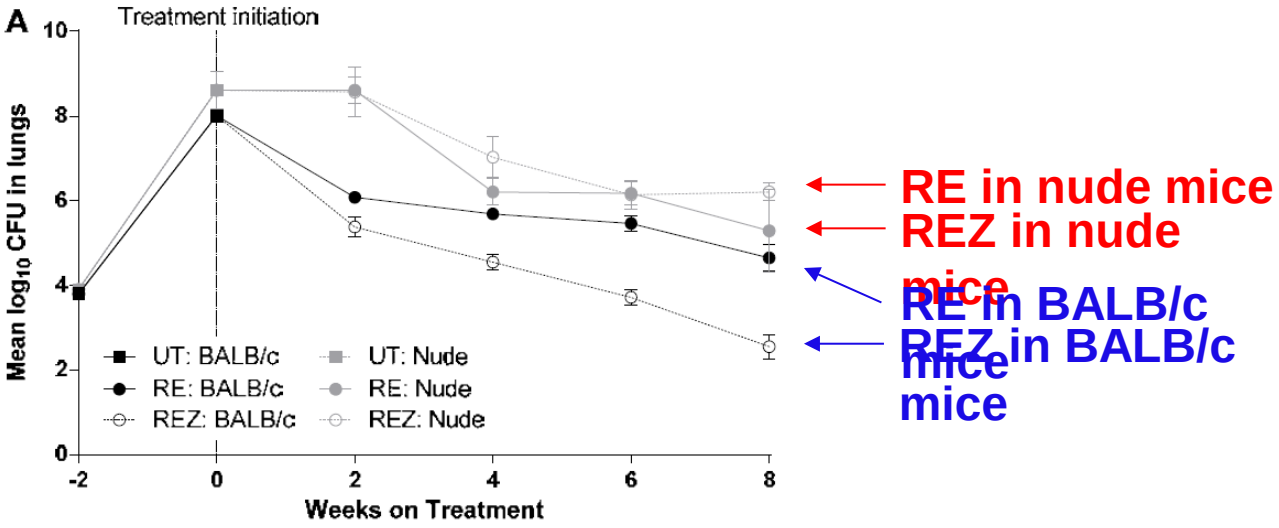
Rosenthal et al, AAC 2012; 56:4331

PZA PK/PD in non-clinical models

- AUC/MIC correlates best with activity[1]
- AUC associated with a -0.11 CFU/day reduction:
  - Hollow fiber system = 1500 µg-h/ml[1]
  - BALB/c mouse = 323 µg-h/ml[2]
- More potent effect of PZA in mice is likely due to lower pH (≤ 5) inside mature phagosomes of activated macrophages[3] vs. that in the HFS-TB (5.8) which effectively reduces the PZA MIC by ~10x
- Increasing current dose by 2-4x increases kill rate[1,2]

[1] Gumbo et al, AAC (2009); 53:3197
[2] Lanoix et al, AAC (2016); 60:735
[3] Vandal et al, Nat Med 2008; 14:849

PZA (Z) is relatively ineffective in immunocompromised mice



RE in nude mice

REZ in nude mice

RE in BALB/c mice

REZ in BALB/c mice

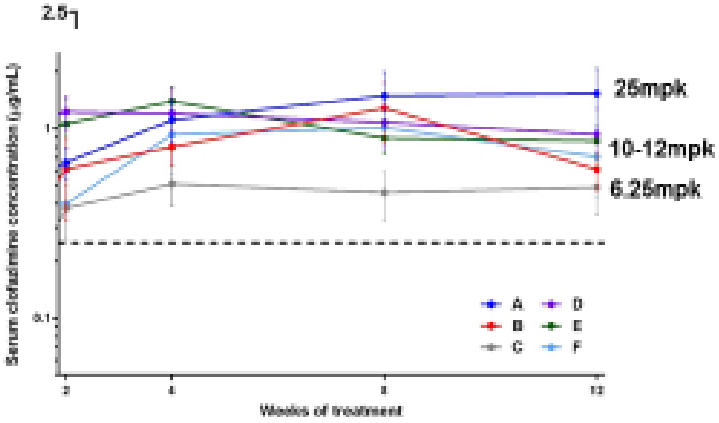
Almeida et al, Mycobact Dis 2014; 4:145

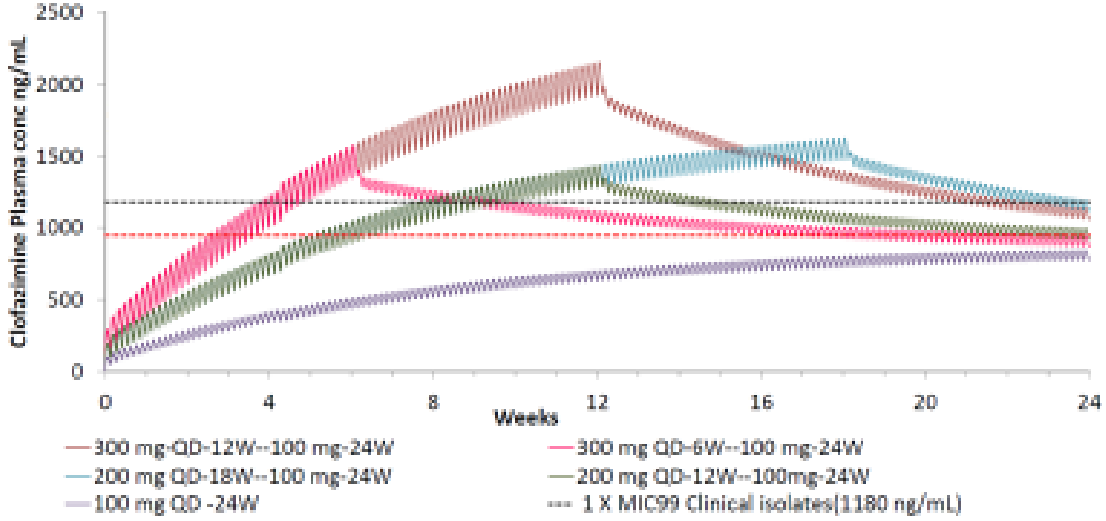
Clofazimine PK: mouse vs. human dose equivalence

Mouse

Mouse	Human
20 mg/kg =	~200 mg
10 mg/kg =	~100 mg

Human

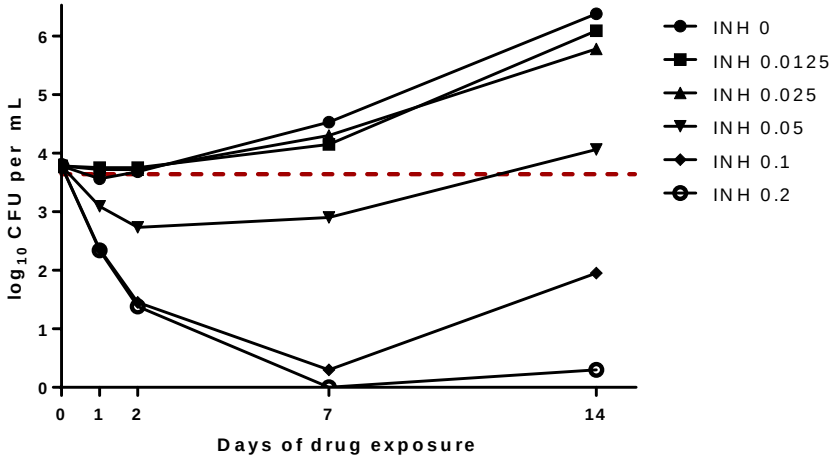


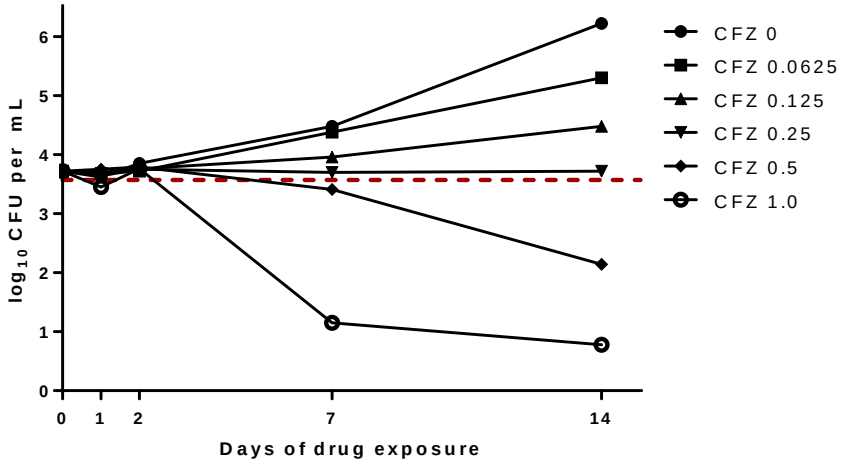


Swanson et al, AAC 2015; 59:3042

From Ganesan S, Sunkara G, McNeeley D and Hughes D. Novartis. UNION Congress. Cape Town 2015.

In vitro EBA of INH and CFZ at similar multiples of their MICs





- MICs: INH = 0.05 µg/mL, CFZ = 0.25 µg/mL

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