

ABSTRACT

Background: Current therapy for pulmonary *Mycobacterium avium*-complex [MAC] disease achieves poor sustained sputum conversion rates and is poorly tolerated. SPR719, the active moiety of SPR720, a novel gyrase inhibitor, has demonstrated low MICs against MAC. SPR720 is being developed as an oral therapy for use in combination with other antibiotics for the treatment of patients with pulmonary disease due to infection with MAC.

Objectives: To identify SPR719 pharmacokinetic/pharmacodynamic [PK/PD] parameters and optimal SPR720 dose for treatment of pulmonary MAC.

Methods: SPR719 was administered once daily for 28 days using the human half-life of 3.3 hours in the hollow fiber system model of pulmonary intracellular MAC [HFS-MAC]. Bacterial burden, including for SPR719-resistant subpopulations, and drug concentrations, were measured via repetitive sampling of HFS-MAC units. A separate dose fractionation study in the HFS-MAC was used to identify the PK/PD index linked to effect. MAC burden versus SPR719 exposure was modeled using the inhibitory sigmoid maximal effect [E_{max}] model and resistance using the “antibiotic resistance arrow of time” model. Finally, we performed preliminary Monte Carlo Experiments to guide the clinical dose selection for SPR720 monotherapy.

Results: The median HFS-MAC intracellular-to-extracellular SPR719 AUC_{0-24} ratio was 1211:1. The PK/PD parameter best linked to microbial kill was determined to be AUC_{0-24}/MIC . SPR719 E_{max} was $-1.5 \log_{10}$ cfu/mL compared to day 0; $1.0 \log_{10}$ cfu/mL reduction and acquired-resistance suppression were achieved by an AUC_{0-24}/MIC of 2.0 and 11, respectively. SPR720 1,000 mg/day monotherapy was predicted to achieve $1.0 \log_{10}$ cfu/mL kill in 95%, and resistance suppression in 43%, of 10,000 simulated subjects.

Conclusions: SPR720 monotherapy is predicted to achieve exposures associated with bactericidal effect against pulmonary MAC in 95% of patients at doses that have recently been established to be safe and well tolerated. These data support the continued development of SPR720 for the treatment of MAC-PD.

METHODS, MODELING & SIMULATIONS

Materials

- Bacterial isolate.** *Mycobacterium avium* (ATCC 700898) and THP-1 cells (ATCC TIB-202) were purchased from ATCC (Manassas, VA).
- Drug.** SPR719 was provided by Spero Therapeutics, Cambridge, MA.

Minimum Inhibitory Concentration

- MIC studies were performed using broth dilution method using cation-adjusted Mueller Hinton broth [CAMBH] + 5% OADC and was 1 mg/L.

SPR719 Exposure-Effect Study in HFS- MAC

- 20mL of MAC infected THP-1 monocytes were inoculated into the peripheral compartment of each HFS-MAC unit, which had circulating RPMI and 2% FBS.
- SPR719 was administered to HFS-MAC units in duplicate, to achieve 0-24hr area under the concentration-time curves [AUC_{0-24}] of 0, 0.72, 1.56, 1.92, 1.99, 2.15, 10.84 and 117.55 mg-h/L. We mimicked a human serum half-life of 3.3h. The concentrations achieved in the external compartment and inside infected macrophages were validated by direct measurement [Deshpande et al., AAC 2016].
- The peripheral compartment was sampled on days 0, 7, 14, 21 and 28.
- THP-1 cells were ruptured, and bacteria cultured on Middlebrook 7H10 agar for colony forming units counts, as well as on agar supplemented with 1.5 times the SPR719 MIC to capture drug-resistant sub-population.

Dose-Fractionation Study in HFS-MAC

- 0, EC_{10} , EC_{20} , EC_{50} , EC_{80} and EC_{90} exposures identified in the dose effect study were administered to HFS-MAC units.
- All doses were administered daily. In addition, for the EC_{20} , EC_{50} , and EC_{80} , the daily dose was either split into two and administered every 12 hours or two daily doses were combined and administered every other day.
- Sampling procedures were as in the dose-effect studies.

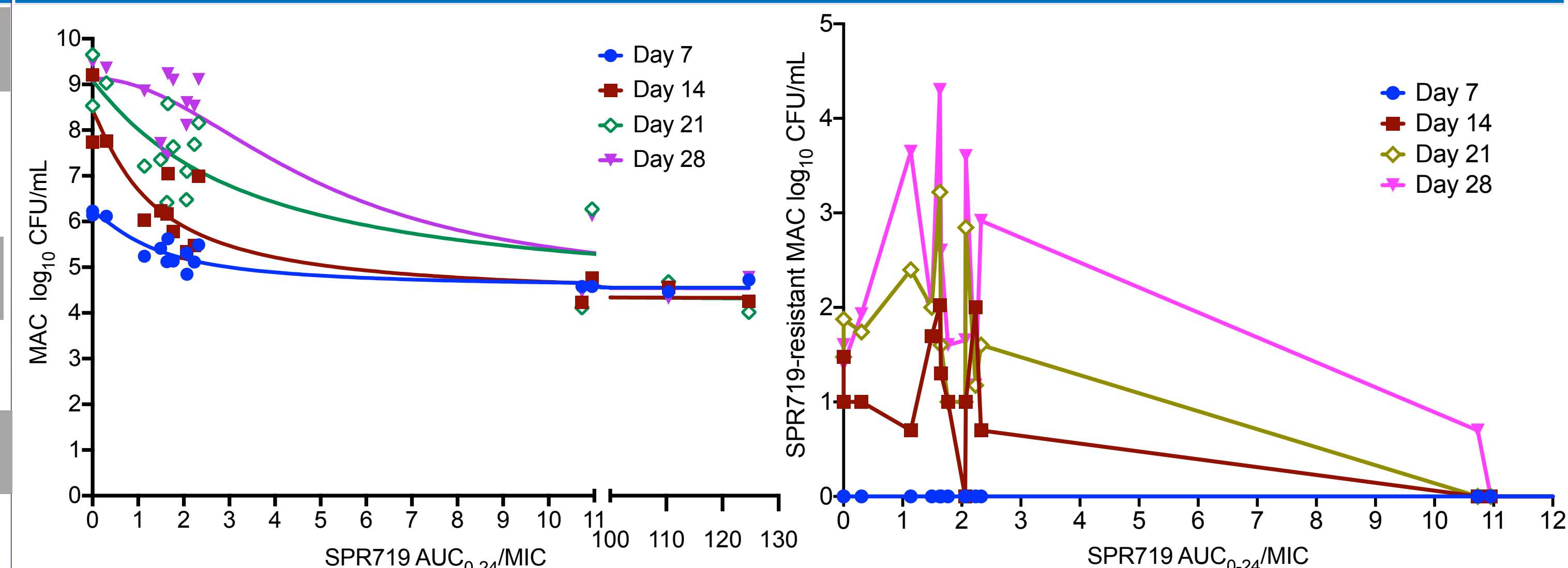
PK/PD Analyses: Microbial Kill & Resistance

- The relationship between total bacterial burden and SPR719 AUC_{0-24}/MIC ratio was examined using the inhibitory sigmoid E_{max} model.
- The relationship between SPR719-resistant population and AUC_{0-24}/MIC was modeled using the “antibiotic resistance arrow of time”-based quadratic function

Monte Carlo Experiments

- Implemented using ADAPT 5 [Biomedical Simulations Resources], for 10,000 patients
- Domain of input in subroutine PRIOR of ADAPT 5: Mean clearance [%CV] of 60.2 L/hr [41.6%], Volume of 292 L [37.2%], and K_a of $0.223/hr^{-1}$ from Spero human PK study
- Penetration of SPR719 into lung based on epithelial lining fluid AUC penetration ratio of 0.413 from non-human primate BAL study
- Doses of 500, 600, 700, 800, 1000, and 1,500 mg/day were modeled for their ability to achieve the AUC/MIC targets of: [1] $1.0 \log_{10}$ CFU/mL kill compared to day 0, and [2] exposure associated with resistance suppression.
- MIC distribution based on the literature [Brown-Elliott B, et al. Antimicrob Agents Chemother 62, 2018].

HFS-MAC EXPOSURE-EFFECT RESULTS



- SPR719 mediating $1 \log_{10}$ cfu/mL kill below day 0 burden was AUC/MIC of 2.0 ± 0.51
- SPR719 mediating EC_{80} was AUC/MIC of 4.09 ± 1.04
- SPR719 associated with resistance suppression was AUC/MIC of 11

HFS-MAC DOSE FRACTIONATION RESULTS

Akaike Information Criteria Scores

Sampling day	Day 7	Day 14	Day 21	Day 28
% time above MIC	Not converged	Not converged	Not converged	Not converged
C_{max}/MIC	-33.70	-7.631	9.333	2.577
AUC/MIC	-36.48	-10.85	-5.844	-6.767

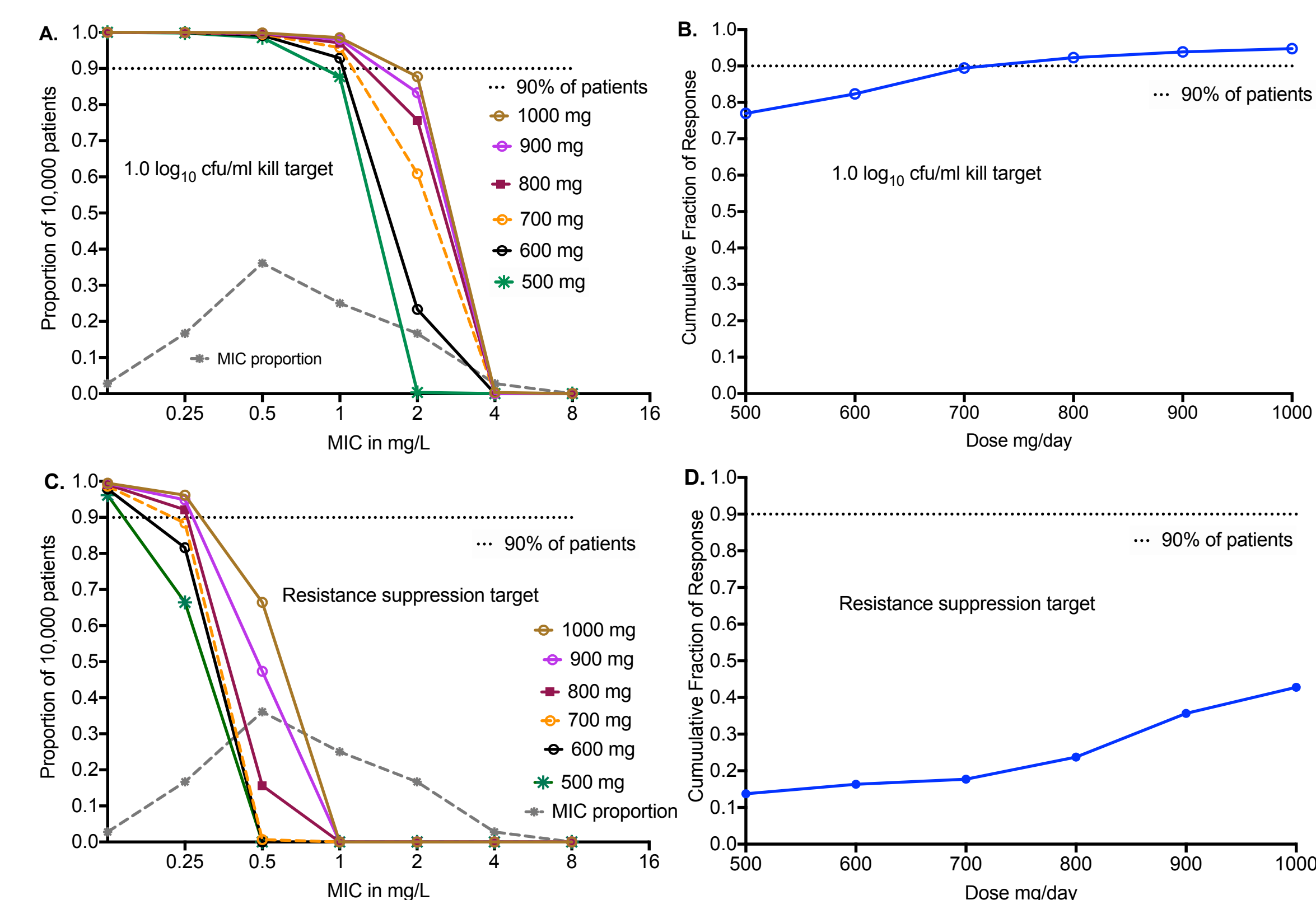
BACKGROUND AND RATIONALE

- Pulmonary *Mycobacterium avium* complex [MAC] disease is one of the more difficult diseases to treat. The recommended combination therapy regimen of a macrolide [either clarithromycin or azithromycin], ethambutol, and rifabutin, is associated with a sustained sputum conversion rate of only 54% (Pasipanodya J et al, J Antimicrob Chemother 2017)
- The standard regimen is poorly tolerated by patients based on patient surveys, and macrolide-based discontinuation rates are encountered in up to 25% of patients (Kwon YS et al, J Antimicrob Resist 2020). In addition, the average duration of macrolide-containing therapy is 18 months, a long time to be on high adverse event therapy with poor sputum conversion rates ((Pasipanodya J et al, J Antimicrob Chemother 2017; Kwon YS et al, J Antimicrob Resist 2020). Thus, there is an immense unmet need for new type of drugs and combinations that could be better tolerated
- SPR719, is the active moiety of the prodrug SPR720, and a novel aminobenzimidazole. SPR720 has the novel mechanism of action of inhibiting the ATPase activity of gyrase in mycobacteria, a target site different from that of quinolones
- A recent study identified the SPR719 minimum inhibitory concentrations [MICs] range of 0.12- 2mg/L, and the MIC to inhibit the growth of 50% [MIC_{50}] and 90% [MIC_{90}] of clinical M. avium and MAC-X isolates of 0.5/2.0 mg/L, and 1/1 mg/L, respectively (Brown-Elliott BA et al, Antimicrob Agents Chemother 2018).

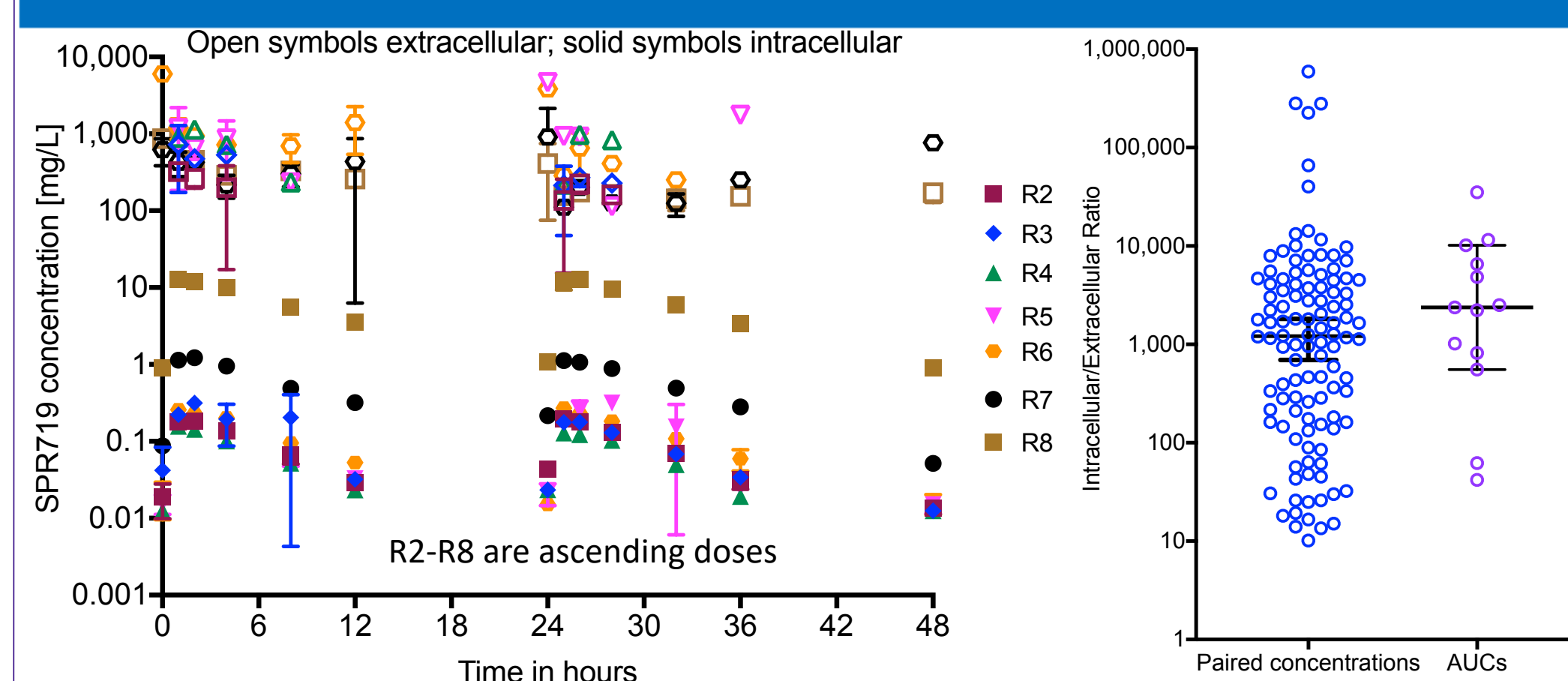
Pulmonary MAC is a predominantly intracellular pathogen in monocyte-lineage cells in alveoli and inside multinucleated giant cells in necrotic lesions (Hibiya K et al, Pathol Res Pract 2012).

- Here we used the hollow fiber system model of MAC [HFS-MAC], a preclinical pharmacokinetic/pharmacodynamic [PK/PD] model in which monocyte-lineage cells are infected with MAC (Deshpande et al, J Antimicrob Chemother 2017) to perform SPR719 exposure-effect studies, followed by dose fractionation studies
- HFS-MAC-derived PK/PD parameters for $1.0 \log_{10}$ CFU/mL kill and resistance suppression were then used as target exposures, by taking into account SPR720 population pharmacokinetic parameter estimates and variance, and drug penetration into lungs, to estimate potential doses for use in the clinic.

10,000-patient MONTE CARLO EXPERIMENT RESULTS



RESULTS: HFS-MAC PKs



DISCUSSION

- SPR719 is associated with bactericidal effect [$1 \log_{10}$ cfu/mL kill below day 0 burden] in the HFS-MAC.
- The efficacy of SPR719 is greater than that of currently recommended drugs as monotherapy in the same HFS-MAC model [Deshpande et al. AAC 2010; Schmalstieg et al AAC 2012]
- MAC is predominantly an intracellular pathogen, the high penetration achieved by SPR719 in infected THP-1 cells is advantageous for MAC therapy.
- SPR720 doses of 750-1,000 mg QD predicted to achieve exposures associated with bactericidal effect in 95% of patients.