

Informativeness of different *in vitro* *Mycobacterium tuberculosis* culture phases in time-kill assay for pharmacokinetic-pharmacodynamic translation

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Background and objectives

Performing time-kill assay (TKA) experiments can be lengthy, depending on the bacterial growth phase being measured. The stationary-phase is when the bacteria is in equilibrium and mimics the human infection more at the time of treatment. However, the experiments require more time than the log-phase culture where the bacteria is exponentially growing.

The aims were to evaluate:

- the identifiability of pharmacokinetic-pharmacodynamic (PKPD) relationships using only log-phase versus combined log- and stationary-phase *in vitro* data
- how early bactericidal activity (EBA) predictions from these datasets align with observed clinical EBA outcomes.

Methods

- In vitro* TKA data with *M. tuberculosis* H37Rv in log- and stationary-phase, in the absence of drug or in the presence of rifampicin or isoniazid, were used to estimate the PKPD relationships of rifampicin and isoniazid as monotherapy [1].
- The final PKPD models were integrated with translational factors to predict EBA simulations [2].
- The EBA simulations were then compared to the observed clinical EBA data.
- The *in vitro* data were modeled in NONMEM 7.5.1 and EBA predictions were simulated using the “deSolve” (version 1.4) package in R (version 4.2.3).

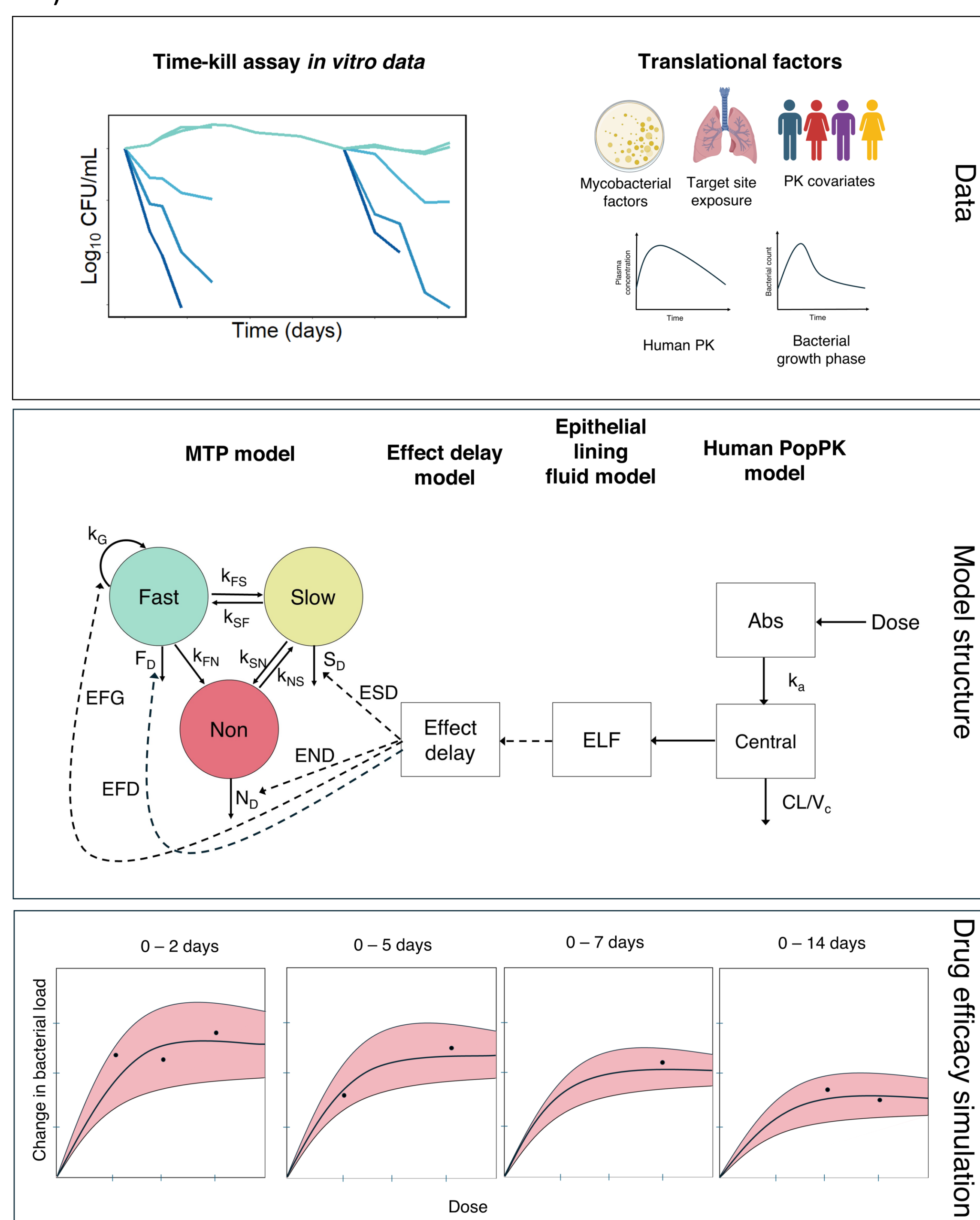


Figure 1. Schematic figure of the modeling workflow

Acknowledgments

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Results

- PKPD modelling using only log-phase data showed rifampicin inhibited the growth of fast-multiplying (F) subpopulation and killed both F and slow-multiplying (S) subpopulations.
- Including the stationary-phase revealed an additional killing effect on non-multiplying (N) subpopulation and improved alignment of EBA predictions with clinical data.

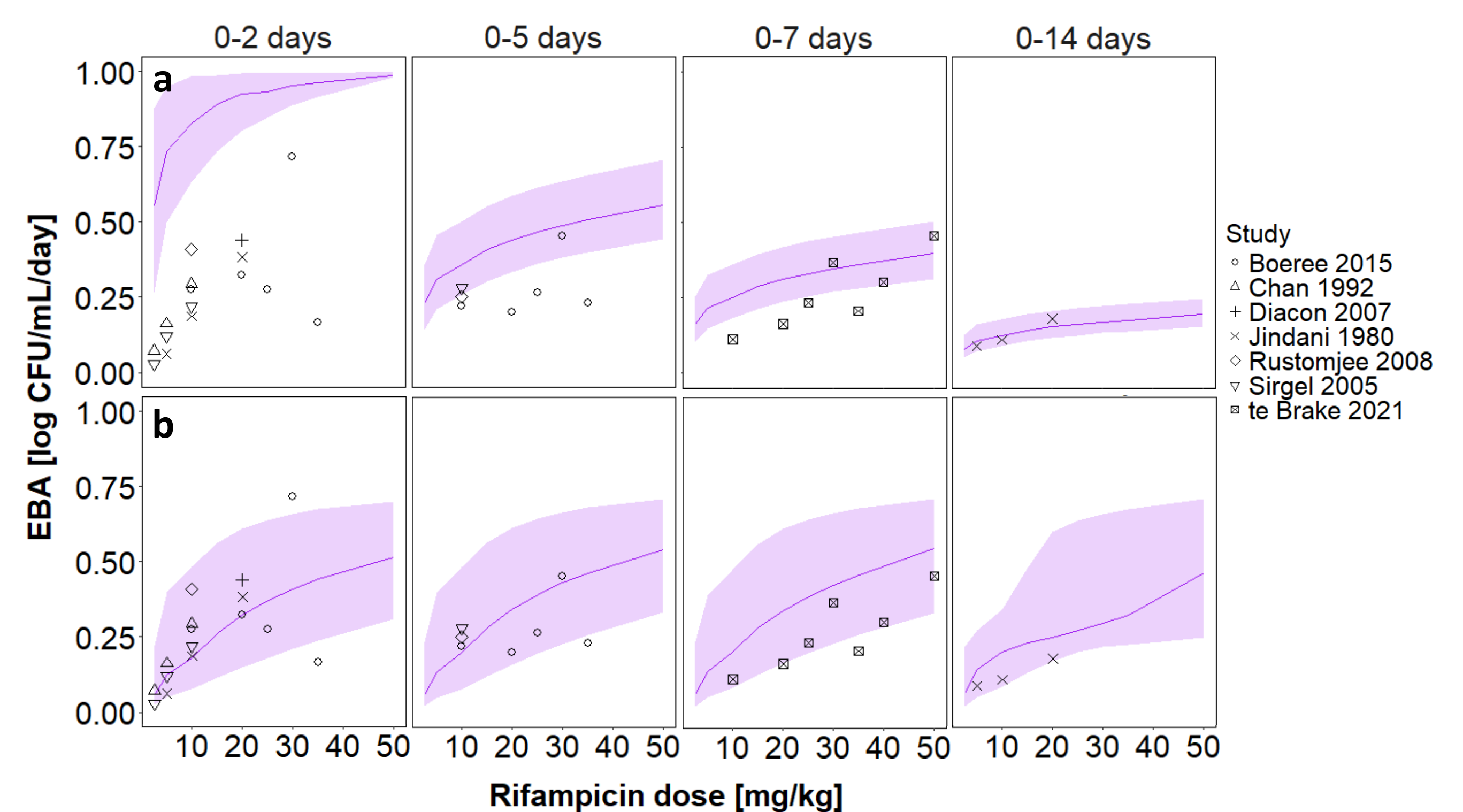


Figure 2. EBA predictions of rifampicin using a) log-phase and b) log- and stationary-phase time-kill assay data

- PKPD effects included inhibition of F growth and killing of both F and S subpopulations, with consistent outcomes whether using log-phase only or combined log- and stationary-phase data.
- EBA predictions for isoniazid were largely consistent with observed data across different studies, irrespective of the use of *in vitro* TKA data.

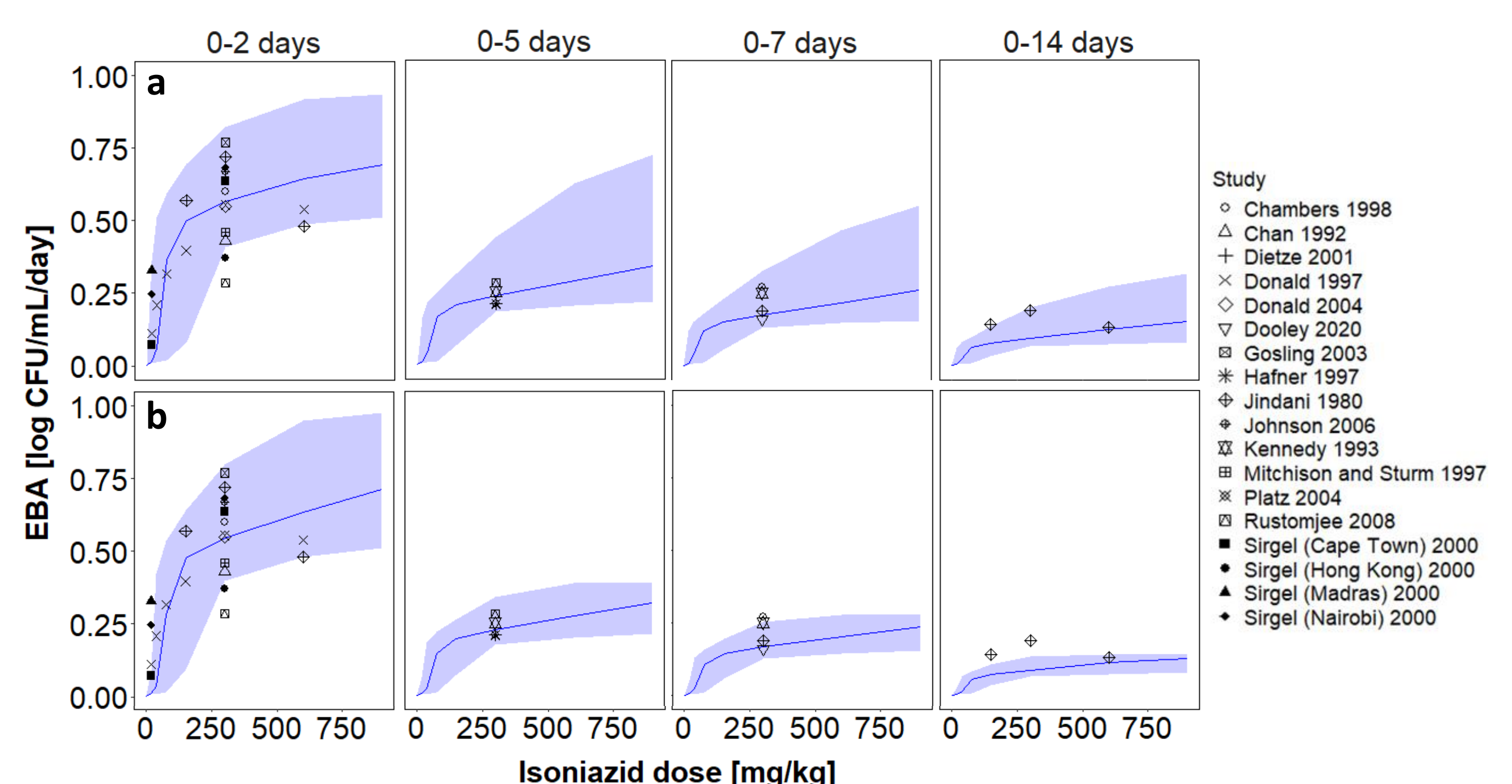


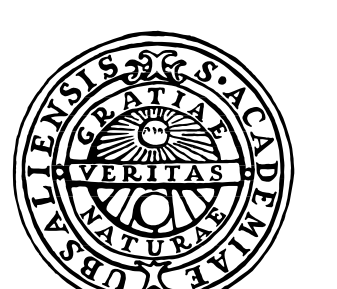
Figure 3. EBA predictions of isoniazid using a) log-phase and b) log- and stationary-phase time-kill assay data

Conclusions

- Including both log- and stationary-phase TKA data is unnecessary for EBA predictions if the antitubercular drug does not affect non-multiplying bacteria.
- However, for drugs that target non-multiplying bacteria, incorporating both log- and stationary-phase data is essential for accurate early clinical efficacy predictions.

References

- [1] O. Clewe et al. *J. Antimicrob. Chemother.*, (2016)
- [2] S. G. Wicha et al. *Clin. Pharmacol. Ther.*, (2018)



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