Implementation of the rRNA burden and rRNA synthesis ratio biomarkers within ERA4TB

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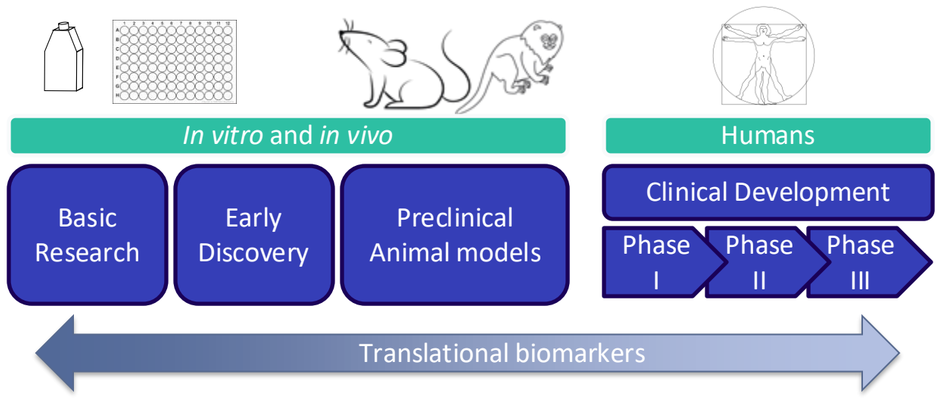
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FACTS & FIGURES

Start date:	01/01/2020
End date:	31/12/2026
IMI funding:	89 815 600 €
EFPIA in kind:	118 148 291 €
Total Cost:	207 963 891 €
Project website:	https://era4tb.org/
Social media:	https://www.linkedin.com/company/era4tb/

CHALLENGE

Throughout the process of drug discovery and treatment development, multiple efforts are made at the different stages, from initial *in vitro* assays to the clinical development. Ideally, collection of comparable information across these stages, could enhance translational insights, thereby, establishing connections and potentially predicting behaviours at later stages.

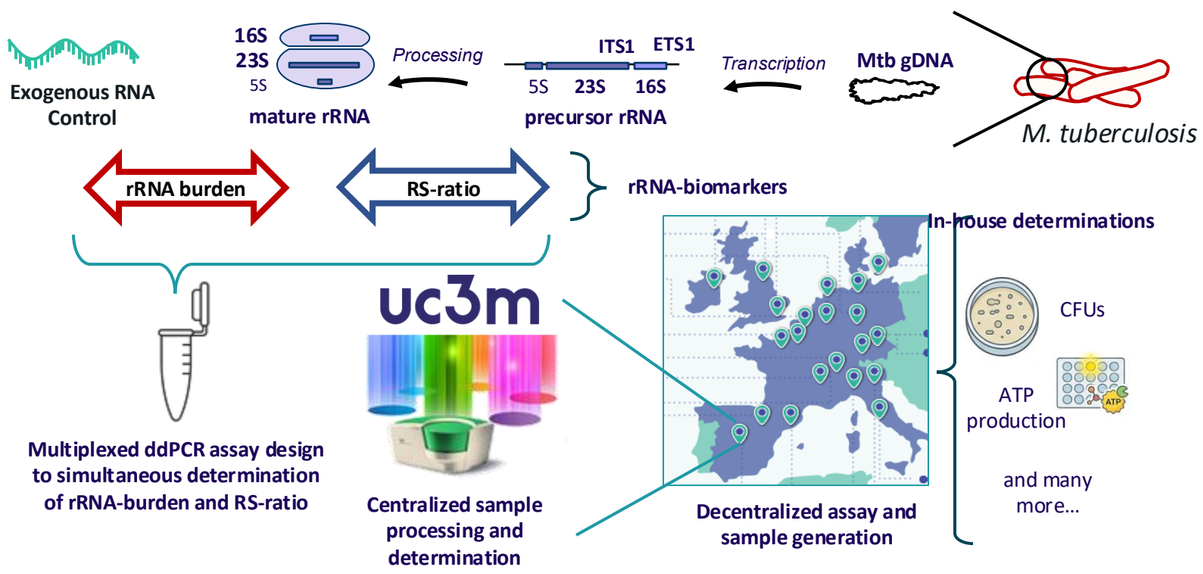


In tuberculosis, rRNA-based biomarkers have been recently implemented with special emphasis in animal and clinical stages, as surrogates of bacterial burden (MBLA & **rRNA burden**) and metabolic state of the bacteria (rRNA synthesis/**RS-ratio**).

We propose the implementation of the rRNA-based biomarkers from the earliest stages of ERA4TB, where massive *in vitro* efforts are being made, to compare with later *in vivo* stages and feed models for translational characterization of the treatment development.

APPROACH & METHODOLOGY

Within ERA4TB we implemented molecular translational biomarkers based on the quantification by droplet digital PCR of rRNA components.

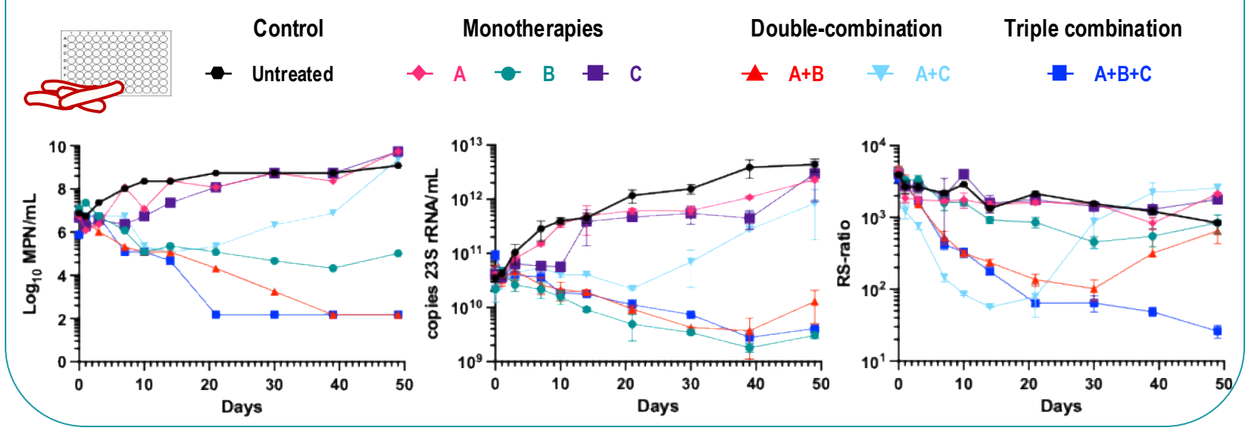


Our workflow allows decentralize sample generation of *in vitro* or *in vivo* BSL3 platforms across Europe, and centralize the processing and simultaneous determination of both biomarkers in a multiplexed assay, to complement in-house determinations.

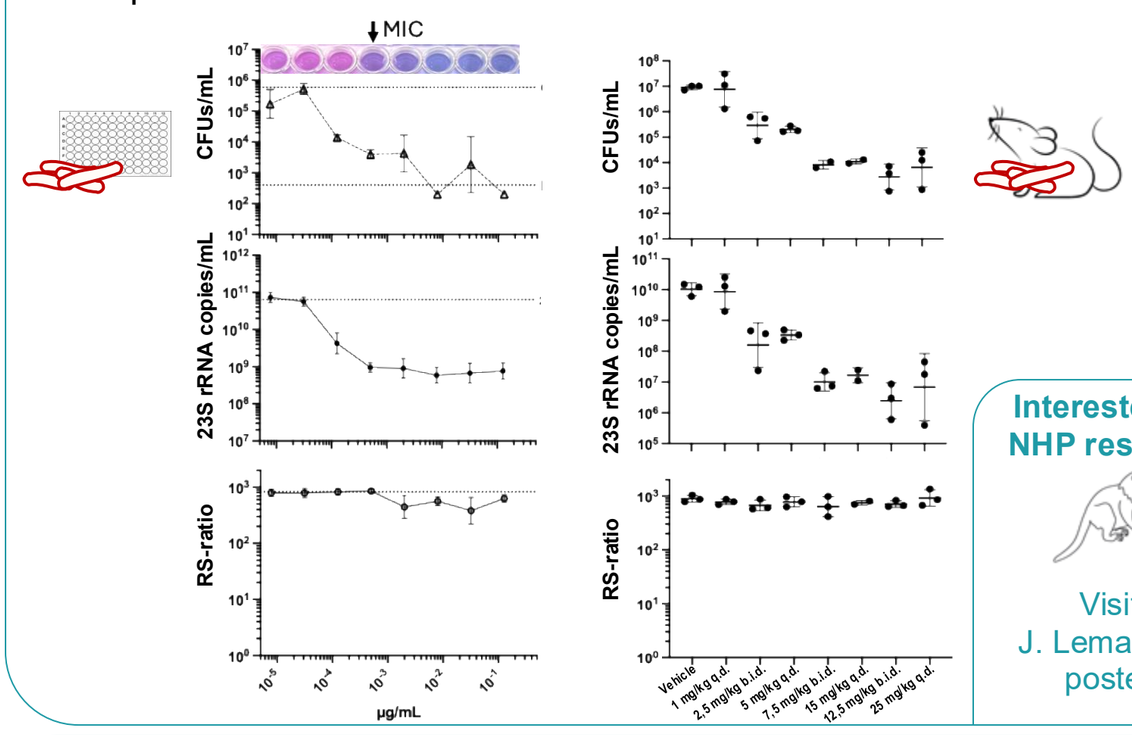
We are exploring these biomarkers in other pathogens — *M. ulcerans*

RESULTS

Mtb in vitro time kill assay reveals how three different drugs act alone or in combination in either culturable bacteria (MPN), rRNA burden and RS-ratio, highlighting the complementary nature of the biomarkers from the same wells



Different concentrations of one asset tested *in vitro* and *in vivo* as an example of how we can obtain translational information

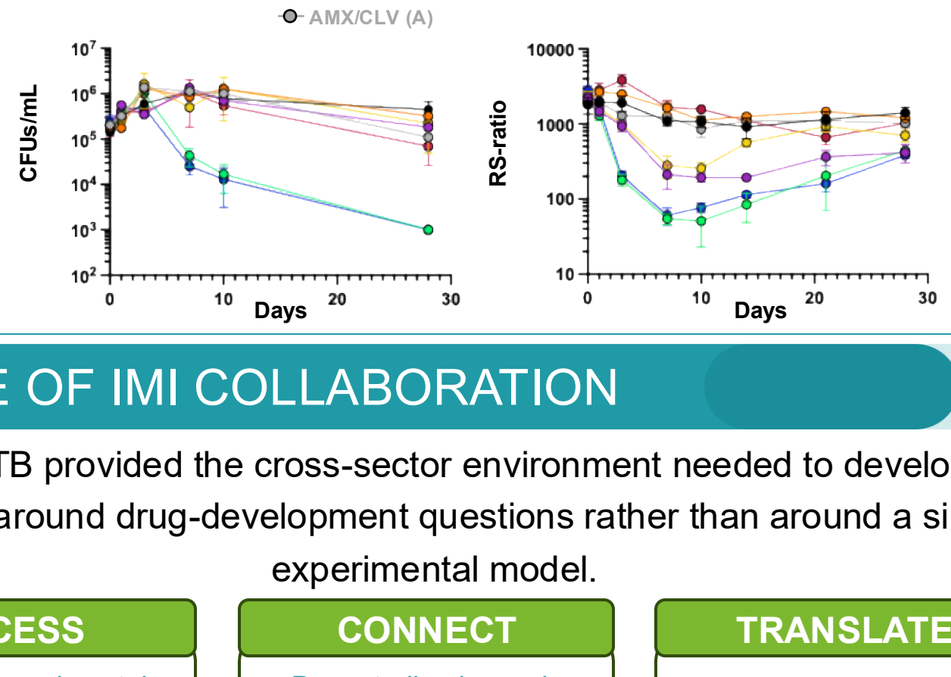


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RS-ratio was successfully implemented for *M. ulcerans* and used to retrospectively characterize drug combinations currently in clinical trials.

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VALUE OF IMI COLLABORATION

IMI/ERA4TB provided the cross-sector environment needed to develop this platform around drug-development questions rather than around a single experimental model.

ACCESS
Different experimental models, drug studies and expertise across European partners.

CONNECT
Decentralized sample generation with centralized, standardized molecular analysis.

TRANSLATE
Same rRNA biomarkers compared across experimental settings.

CAREER IMPACT

WHAT I LEARNED

To think beyond the assay: biological meaning, experimental context and translational relevance matter as much as analytical performance.

SKILLS I DEVELOPED

Molecular assay implementation · biomarker interpretation · collaborative research across experimental platforms · local project coordination

HOW ERA4TB HAS SHAPED MY CAREER

Built a European scientific network and opened new research directions for rRNA-based biomarkers beyond a single pathogen or model.

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